LONG ISLAND BIOLOGICAL ASSOCIATION

ANNUAL REPORT

OF

THE BIOLOGICAL LABORATORY

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LONG ISLAND BIOLOGICAL ASSOCIATION

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ANNUAL REPORT

OF

THE BIOLOGICAL LABORATORY

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FIFTY-NINTH YEAR

1948

TABLE OF CONTENTS

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REPORT OF THE DIRECTOR

The purpose of the Biological Laboratory, when it was organized almost sixty years ago, was to provide summer research facilities for scientists who desired to work at the seashore, and to offer courses in marine biology and other biological subjects for college students. During the first period of its existence the Laboratory functioned exclusively as a summer institution. When it was reorganized in 1924, a full-time research program was added to the other activities, and as a result the Laboratory began to function all year. Through the generosity of Mr. and Mrs. Acosta Nichols and Mrs. Walter B. James, two laboratories were built to take care of these new needs. After a second reorganization, in 1941, when the program was coordinated with that of the Department of Genetics of the Carnegie Institution, the emphasis on research became still stronger. It took several years, however, to develop a well-rounded research program, particularly since the war brought interruptions to the normal course of progress. At the present time it may be said that our goal has been reached. The Laboratory now has a well-defined research program, which is self-contained but which derives much support and strength from its coordination with the research program of the Department of Genetics.

The current program of the Laboratory includes two projects. The first, which has been developing for the past three years, consists of genetical, biochemical, and physiological studies of the origin of bacterial resistance. Particular emphasis is placed on resistance to bacteriophages, to antibiotics (penicillin, streptomycin, aureomycin, chloromycetin), and to certain specific chemicals. A search is made for chemicals that are capable of inducing hereditary changes in bacteria. This project offers fascinating possibilities for combining the investigation of basic genetical problems with studies that are important for understanding the clinical action of antibiotic substances. Financial support for this project is derived from grants of the Jane Coffin Childs Memorial Fund, the National Tuberculosis Association, and the Army Chemical Corps.

The second research project of the Laboratory is just being organized as this Report goes to press; it will comprise a study of the hereditary effects induced in populations exposed to continuous irradiation. During the past decade considerable knowledge has been accumulated about the hereditary changes that may be induced in individuals exposed to various radiations—particularly X-rays and radium rays. Such changes are transmitted to the offspring of the individuals exposed. However, very little is known about how a group of individuals—a population—would be affected if exposed to prolonged irradiation. It is of particular interest to find out to what extent hereditary abnormalities (which are the hereditary changes most frequently produced) would accumulate in such a population. This knowledge is of special importance at present, when the use of atomic energy has tremendously increased the chances of exposure to radiations. For our experiments we will use the vinegar fly (Drosophila)

because methods are available for conducting such research with this organism. The project is being financed by a grant received from the Atomic Energy Commission.

During the past year research on the genetics of antibiotic formation was carried on with the support of a grant from Schenley Laboratories, Inc. Work on this project is being concluded, since the funds available for it have been exhausted.

In addition to the research program, the Laboratory engaged in its usual summer functions in 1948. The thirteenth Cold Spring Harbor Symposium on Quantitative Biology was held early in June. Later, the course on methods used in research with bacterial viruses was given for advanced scientists, and the course in Nature Study for young people of the community. The summer accommodations of the Laboratory were filled to capacity by visiting scientists.

Research

A very significant discovery in the field of biology was made during the past year by Dr. A. Kelner, research bacteriologist working on the antibiotic project. It is known that if microorganisms are exposed to ultraviolet rays many of them are killed, the number depending on the intensity of the radiation and the length of exposure. About seven years ago it was reported at our Symposium that if such "killed" microorganisms are kept for a while some of them become alive again—that is, they recover. This recovery was seen to vary from experiment to experiment, and the cause of it is not known. Kelner, who has made extensive use of ultraviolet irradiation in his experiments with fungi, actinomycetes, and bacteria, became interested in this problem and succeeded in solving it. He found that recovery takes place if the irradiated organisms are exposed to visible light. Apparently the chemical reaction which is induced in cells by ultraviolet radiation, and which is responsible for their death, can be either reversed or neutralized by subsequent exposure to strong visible light. This discovery has furnished a new lead for the study of biological effects of radiations, and may help toward a better understanding of the processes occurring in living cells.

Dr. Vernon Bryson, our biologist, found that ultraviolet irradiation of the medium in which bacteria are cultured will either retard or suppress bacterial growth. Presumably, some substance injurious to bacteria is induced in the medium by the treatment. Experiments indicate that this substance may be either identical with or related to peroxide. It is interesting to note that stranis of bacteria that are resistant to radiations are less affected by this substance than are radiation-sensitive strains, which suggests some relationship between bacterial resistance to redation and the action of the substance induced by irradiation.

Bryson is experimenting also with the bacillus responsible for tuberculosis in animals, and has found that complete resistance to streptomycin in this bacillus originates through a hereditary change that occurs approx-

imately once per one billion cell divisions. Our chemist, Dr. Bo Prytz, is trying to find out whether or not the change resulting in resistance can be traced to a change in some measurable chemical property of bacterial cells. This work is still in an early stage.

The nature of summer research at the Laboratory depends on the group of scientists who are there during any particular season. In 1948 there was a good representation of scientists interested in research with microorganisms. A great majority of these worked with bacterial viruses (bacteriophages). Professor Mark H. Adams, of the New York University College of Medicine, who was in charge of the Course on Bacteriophages, investigated the role of calcium ions in the life processes of a phage known as T5, and established that calcium is necessary not only for multiplication but also for survival of this phage. Dr. G. S. Stent, of the California Institute of Technology, took the phage course during the early part of the summer and continued with research until the end of the season. His anaylsis of the kinetics of the virus-antiserum reaction supports the assumption that active phage becomes progressively more difficult to inactivate with prolonged exposure to antiserum.

Dr. S. S. Cohen, of the University of Pennsylvania, developed a method for quantitative analysis of sugar phosphates by paper chromatography. He also investigated the correlation between increase in desoxyribose nucleic acid and phage synthesis.

Professor S. E. Luria and Dr. R. Dulbecco, of Indiana University, made crosses between several phage mutant and inactivated types obtained by ultraviolet irradiation. They concluded that in T2 phage there are at least twenty-five independently transferable loci at which lethal mutations may be produced.

Mr. J. D. Watson, graduate student from Indiana University, studied the reactivation of X-rayed phage, and concluded that the probability of reactivation and of genetic transfer is much less in X-ray-inactivated phabe than in phage inactivated by ultraviolet rays.

Dr. A. W. Bernheimer, of New York University College of Medicine, made an unsuccessful search, in material collected from several ponds in Nassau County, for viruses affecting protozoa.

Miss Margaret Lieb, graduate student from Columbia University, made a preliminary study of the mutagenic activity of several chemicals on the bacterium Escherichia coli.

Dr. L. Michaelis, of the Rockefeller Institute for Medical Research, continued with studies made the previous summer on the staining properties of fixed sections with respect to acid dyes. His associates, Dr. S. Granick and Dr. S. H. Wollman, continued with the analysis of various components of chlorophyll pigment.

Dr. V. Menkin, of Temple University School of Medicine, studied a toxic substance released from severely injured vertebrate cells, which he has named "necrosin". He found that crushed tissues of either the soft-shell clam or the horseshoe crab yield an extract having the same properties as necrosin.

Dr. Harold A. Abramson carried on research on several basic problems relating to allergies. In particular, he studied the behavior of hydrogan peroxide in the skin, and experimented with the electrophoretic fractionation of ragweed pollen.

Several other scientists used their time at the Laboratory for writing. Dr. Ernst Mayr, of the American Museum of Natural History, worked on a book which will be called "The Biology of Birds." Dr. W. H. Sheldon, of the College of Physicians and Surgeons—author of "The Varieties of Human Physique" and "The Varieties of Temperament"—finished writing and revising his new book "Varieties of Delinquent Youth," and did preliminary work on an "Atlas for Somatotyping Men."

Brief statements by the investigators about their research at the Laboratory are presented in the section "Reports of Summer Investigators."

Symposium

The series of conferences known as the Cold Spring Harbor Symposia on Quantitative Biology was started in 1933 and has been held every summer since then except for three years during the war. The aim of these conferences is to bring together scientists working on different aspects of a certain problem, and to give them an opportunity of discussing their worok at leisure in the congenial environment ment of the Laboratory.

The 1948 symposium was held June 8 through June 16, on the subject "Biological Applications of Tracer Elements." Tracer elements are now used intensively in biological research. More than twenty years ago it was discovered that some of the well-known elements exist in several different forms; and that some of these forms, because they are radioactive or because of unusual atomic weight, can be readily detected—or "traced"—in various chemical or biological processes. Therefore, if a scientist wants to study the function of a chemical compound in a life process, he can add to it one of these tracer elements, and feed or inject the mixture into the living organism he wishes to study. Then, by certain physical methods that have been developed, he can determine the distribution of the tracer substance within the tissues of the body, and thus follow the path of the chemical. In this way physiological processes such as metabolism are being made clearer, through work on a number of different organisms.

Atomic energy research has recently provided the means of producing such tracer elements in large quantities, and has made an abundant supply available for research purposes. Consequently many laboratories throughout the United States and in other countries have started extensive programs to investigate a variety of biological and medical problems by this means. Research is now being done on some of the most primitive organisms—like bacteria, yeasts, and fungi—and also on the most complex ones, including man. These different researches may have equal importance; for it is a well-established fact that fundamental discoveries made on any living organism, from the very simplest to the most complex, will have a general application and will be true for all.

Many facets of the various problems being studied were brought out

in the lectures and in the extensive discussions at the meetings. This symposium was one of the first international conferences to discuss the recent results of such work.

The foreign speakers on the program were: Dr. T. J. Arnason, from the University of Saskatchewan, Canada; Dr. Ronald Bentley, National Institute for Medical Research, London, England; Professor Karl Bernhard, University of Zurich, Switzerland; Dr. Gosta Ehrensvard, University of Stockholm, Sweden; Professor Einar Hammarsten, Karolinska Institutet, Stockholm, Sweden; Dr. George Hevesy, University of Stockholm; and Professor Hans H. Ussing, University of Copenhagen, Denmark. Dr. Hevesy was the first scientist to use tracer elements, and for this work he received the Nobel Prize in 1943.

Speakers from this country who presented papers on the program came from many different institutions, including the Argonne, Brookhaven, and Oak Ridge National Laboratories; the Sloan-Kettering Institute for Cancer Research; the College of Physicians and Surgeons; and the Medical Schools of Howard University, Tulane University, Washington University, Western Reserve, and the Universities of California, Chicago, and Pennsylvania. Other registrants at the meetings, numbering more than 163, came from 15 states and from Chile, France, Holland, and Norway.

The lectures and discussions, held in Blackford Hall, occupied most of the visitors' time during the nine-day symposium. One afternoon the group visited the Brookhaven National Laboratory at Upton, Long Island. There they saw an exhibit of research instruments, visited the laboratories of various research workers, and enjoyed a picnic supper with members of the Brookhaven staff.

All papers prepared for the symposium, and much of the material presented in discussions, were published in book form, as Volume XIII of the Cold Spring Harbor Symposia on Quantitative Biology.

Teaching

The course in Nature Study was given again by Dr. and Mrs. J. Southgate Y. Hoyt, of Cornell University. This course is designed to stimulate interest in nature among the younger generation, by showing them how to observe the many interesting plants and animals around them, by teaching them how to answer the questions raised by their observations, and by making them realize that careful and accurate study of the smaller incidents observed by all of us contributes greatly towards expanding our knowledge of natural phenomena. The course was divided into four sections, according to the ages of the pupils, and was attended by fifty local young people. On the afternoon of the closing day a public exhibition was held to demonstrate the activities of the various classes to parents and friends.

For the fourth successive year an intensive three-weeks course was offered in techniques and problems of research with bacterial viruses. It was taught by Professor Mark H. Adams of the New York University

School of Medicine. There was a capacity enrollment of 14 students taking both lectures and laboratory, plus two more who attended only the lectures. In connection with the course, a series of ten seminars was arranged. It is interesting to note that, of the 47 individuals who have taken this course so far, 30 have been active research workers with a doctorate, and most of the remainder have been graduate students working towards an advanced degree.

Lectures

Seminar lectures were held throughout the summer, in cooperation with the Department of Genetics of the Carnegie Institution. The speakers were summer members of the Laboratory, and arrangements were in the charge of Dr. Ernst Caspari of the Department of Genetics. A list of speakers and titles is given below:

June 24: L. Michaelis, Rockefeller Institute. Oxygenation and oxydation; the problem of oxyhemoglobin.

July 1: S. E. Luria, Indiana University. A radiation study in bacteriophage genetics.

July 8: Valy Menkin, Temple University. Mechanisms of inflammation.

July 15: Seymour S. Cohen, University of Pennsylvania. Studies on Rickettsiae and typhus vaccine.

July 22: W. H. Sheldon, College of Physicians and Surgeons. On the natural history of Homo americanus.

July 29: R. B. Roberts, Department of Terrestrial Magnetism, Carnegie Institution. Extrasensory perception.

August 5: Bernard Davis, U. S. Public Health Service, Cornell University School of Medicine. Fats, albumen, and tubercle bacilli.

August 12: S. Granick, Rockefeller Institute. Structure of hemoglobin and functions of some of its parts.

August 19: H. Waelsch, Psychiatric Institute, Columbia University. Studies in glutamic acid metabolism.

August 26: Ernst Mayr, American Museum of Natural History. Species problems.

An illustrated lecture of general interest was given on the evening of September 22, by Dr. Robert Cushman Murphy, Chairman of the Department of Birds of the American Museum of Natural History and President of the Long Island Biological Association. The title was "A Naturalist's Observations in Changing New Zealand," and the lecturer described the very fascinating experiences and impressions of a recent trip to New Zealand. The lecture was well attended by members of the Association and their friends.

Dining Room

The Blackford Hall dining room was in operation from June 7 to September 7, and accommodated both the members of the Biological Laboratory and the resident members of the Department of Genetics. During the

symposium period it served meals to over one hundred persons, and during the remainder of the summer to about fifty persons. Mrs. Ralph Thompson acted as dining-room manager.

Laboratories and Equipment

The research project to study the effects of continuous irradiation on the structure of populations will be housed in the Dr. Walter B. James Memorial Laboratory, and this building is being re-equipped. The facilities will consist of a large general laboratory, a smaller laboratory for cytological studies, an office and study room and an insulated radium-treatment room with temperature control, a constant-temperature culture room, and a kitchen equipped with a washing machine, sterilizer, and cooking facilities.

Several expensive instruments essential for research with bacteria were purchased, with the funds available for that work and with a special grant received from the Brooklyn Cancer Committee of the American Cancer Society. The equipment acquired includes a Sorvall high-capacity centrifuge, a clinical centrifuge, a bacteriological incubator, a Beckman spectrophotometer, and a precision circular Warburg apparatus.

Buildings and Grounds

During 1948 only essential work was done on buildings and grounds. This consisted of exterior painting of Airslie, interior painting of several rooms in residences, building of partitions in Davenport Laboratory to provide for two additional research rooms, and installing of several fluorescent light fixtures in the same building. The heating system in the Dr. Walter B. James Memorial Laboratory has been modernized, and an oil burner installed in the furnace.

Outstanding Needs

The most outstanding need of the Laboratory is an adequate lecture hall. At present the symposium meetings, summer lectures, and conferences are held in the living room of Blackford Hall. This room was not designed for a lecture hall, and therefore its lighting arrangements and other appointments are highly inadequate for the purpose. The room is too small for our needs, and too low-ceilinged for comfort on hot summer days. A new building, with a modern lecture hall accommodating about 150 persons, would give us badly needed facilities.

Moving the lecture room from Blackford Hall would permit the establishment of a comfortable living room there, which would be particularly welcome for rainy days and evenings. New furniture for the living room is needed.

Some of our residence buildings require improvements and repairs. This is particularly true of the dining-room, kitchen, and bathroom facilities in Blackford Hall.

Acknowledgments

It gives me great pleasure to acknowledge the support given to the Laboratory by the members of the Long Island Biological Association. At present only the smaller part of the total expenditures of the Laboratory is covered by the contributions of the membership of the Association; but this part of the budget provides for upkeep and overhead expenses, and is most essential for the existence of the Laboratory.

The Women's Committee, under the presidency of Mrs. George S. Franklin, made an important contribution toward the support of the scientific work of the Laboratory; and its House Committee, under the chairmanship of Mrs. Percy H. Jennings, collected contributions for the furnishing of residences.

Acknowledgment is also made of the contribution of the Wawepex Society toward the upkeep of buildings and grounds, of the John D. Jones Scholarship maintained by that Society, and of a special contribution made this year for repairs in Hooper House.

The Laboratory recognizes with gratitude the research grants made by the Army Chemical Corps, the Atomic Energy Commission, the Jane Coffin Childs Memorial Fund for Medical Research, the National Tuberculosis Association, and Schenley Laboratories, Inc.; the grant made by the Carnegie Corporation, for expenses of foreign guests taking part in the Symposium; and that made by the Brooklyn Cancer Committee of the American Cancer Society, for purchase of scientific instruments.

We wish to acknowledge the assistance given, during the symposium session, by several members of the Brookhaven National Laboratory, and particularly Dr. Leslie F. Nims, director of the Biology Division.

M. Demerec
Director of the Laboratory

REPORTS OF LABORATORY STAFF

V. Bryson, M. Mann, B. Prytz, M. Swanstrom, and E. Yongen

Effect of Chemicals on Bacterial Mutants

Three projects have occupied our attention during the past year. Under a grant from the Jane Coffin Childs Memorial Fund for Medical Research, we have continued the search for chemical mutagens. As reported last year the comparison of carbamates showed that increase in the alkyl chain carbon atoms is directly correlated with rise in toxicity to Escherichia coli. Evidence of mutation was sought in the form of relative increase in the numbers of phage-resistant mutants present in the populations of E. coli exposed to chemical treatment. Such an increase was found. Yet any mutations that might have been induced by methyl, ethyl, propyl, or butyl carbamate could easily have been obscured by a demonstrated incidental selective action favoring the same type of phage-resistant mutants spontaneously present in the population. The occurrence of delayed mutation could not be shown by the phage-aerosol technique of Demerec.

Since urethane (ethyl carbamate) has been stated by other workers to be mutagenic, it was decided to concentrate on this substance and modify the experimental procedure to reveal zero-point mutants if present. The problem was solved very simply. It was reasoned that the relative viability of phage-resistant E. coli compared with the normal strain might vary quantitatively, depending on the strain of bacteriophage used in the test. Utilizing mutants of E. coli that are resistant to phages T1, T4, and T6 (termed B/1, B/4, and B/6, respectively) a comparison was made of the relative viability of these phage-resistant stocks to ethyl carbamate. A strain resistant to a particular phage may also be resistant to other phages, and therefore the cells defined as B/1, B/4, and B/6 are only described in phenotypic terms. Any single category may include several different genetic types having a common property of resistance to a specific phage but otherwise not necessarily similar.

Exposure at 37° C. of a mixed culture of E. coli mutants to 7½ per cent ethyl carbamate solution showed that cells resistant to T1 (B/1) are more resistant to ethyl carbamate than are B/4 or B/6 cells. The comparative viability is shown in the table below.

Relative viability of various phage-resistant strains of E. coli when exposed to 7.5 per cent ethyl carbamate

Time	Total Survivors	B/6	B/4	B/1
0	1.1×10^{10}	5.0×10^{9}	3.1×10^{9}	3.4×10^{9}
30 minutes	1.2×10^{9}	5.2×10^{7}	2.4×10^{6}	9.7×10^{8}
60 minutes	3.3×10^{7}	2.6×10^{6}	2.6×10^{5}	3.5×10^{7}
120 minutes	3.7×10^{4}	0	0	3.5×10^{4}

The column representing the total number of chemically treated survivors at specified times of sampling was obtained by assay of various

dilutions on unphaged nutrient agar. Remaining assays were made on phaged nutrient agar. It is observed that the surviving cells after prolonged exposure to ethyl carbamate must be predominantly the original component of the mixed culture that was resistant to phage T1. The experiment was carefully checked by streaking a minimum of fifty surviving colonies from the various types of agar plates and testing the resistance of each colony to T1, T4, and T6. The resulting streaks were retested for phage resistance pattern. The significant finding in these and subsequent experiments was that selection in favor of B/4 will not occur if mixed cultures of B and B/4 are exposed to ethyl carbamate. When this is done, no evidence of induced mutation with ethyl carbamate can be found at a survival level of 0.9 per cent. At a survival level of 0.006 per cent, however, the number of cells resistant to lysis by T4 shows an increase from 253 per 10^8 to 1.6×10^3 per 10^8. Since routine experiments with laboratory strains of B/4 show that selection by urethane is against the mutant in mixed populations of B/4 and B, it would indeed appear that the requisite protocols had been fulfilled and that mutation of B to B/4 had been chemically induced.

Re-examination of all the facts will show that this has not been proved beyond question. It is known that B/4 colonies may be composed of cells likewise resistant to T3 and T7, or merely to T3. It is also found that so-called B/4 colonies may be morphologically of different types. Potentially, each type of mutant resistant to T4 could differ in its relative sensitivity to a chemical when compared to other T4-insensitive mutants or to the normal strain B. The inclusive and nonspecific nature of the general category B/4 can be shown at high killing levels. If a large number of phage-resistant cells of various mutant types—including laboratory strains of B/1,5, B/4, and B/6—are mixed and exposed to ethyl carbamate, a fraction of the surviving population will be resistant to T4. Subculture reveals that 32 per cent of the B/4 colonies are unable to grow if subcultured, either because lysis is temporarily inhibited in some carbamate-treated cells, or because the resistant colonies represent a type of abnormal bacteria different from the usual mutants designated B/3,4 or B/3,4,7. If many different mutants are resistant to T4, the relative viability of all mutants appearing during experiments designed to test mutation of B to B/4 must be tested against the normal strain. Otherwise, selection may always be involved in any relative increase of B/4 in a chemically treated population of B.

The mutation of E. coli producing a strain resistant to streptomycin and dependent upon it for growth, as described by Miller and Bohnhoff, has provided Demerec with a useful genetic tool to test for chemical induction of mutation. Using this method, we find that the total mutants at a killing level of 0.83 per cent were 124 per 10^8 nondependent on streptomycin. Subtracting the nondependent fraction originally present (21 per 10^8) gives a yield of 103 per 10^8. In a subsequent experiment, at a survival of 1.4 per cent, no significant difference in number of nondependent cells was found, as compared with controls, in a population

treated for one hour with 7½ per cent ethyl carbamate and observed after 72 hours of incubation. Further experiments showed no significant increase in mutant population at the survival level of 2.7 per cent. This negative result is apparent only if mutant colonies are scored after 72 hours of incubation (37° C.). When plates are observed after a week of incubation, the number of mutant colonies able to grow in the absence of streptomycin is nearly twice the 72-hour count. This rise in the number of mutant colonies over a period of several days has been observed before, and may be due either to delayed growth of induced mutants or to the appearance of spontaneous mutants among a bacteriostatically inactivated fraction of the original treated population, which begins growth on the plates after prolonged inactivity. The question may be resolved by making periodic assays of the total number of viable, chemically treated cells present on agar without streptomycin at consecutive periods. It is known that streptomycin-dependent cells may go through a few divisions in the absence of streptomycin. Thus the same difficulty may exist that led other workers to reserve judgment on the final value of reversion in biochemically deficient microorganisms as a test of mutation rate. The final assessment of experiments on the mutagenic effect of carbamates, mercuric chloride, hydrogen peroxide, malachite green oxalate, malachite green hydrochloride, and other chemicals will be made in cooperation with Dr. G. Bertani at the Department of Genetics, who has recently begun a survey of simple chemical substances for mutagenic activity. Our attention for the remainder of this study will be devoted to a complete analysis of the methods.

In a second project, the mutation rate of Mycobacterium ranae, a variety of the tubercle bacillus, has been determined to be 6.4×10^{-9} per bacterium per generation, in the step from streptomycin sensitivity to full resistance at 100 units per ml. The determination was based on the technique first described by Luria and Delbruck, and is contingent on the fact that in a large number of independent populations grown from small inocula of nonmutant cells there is a fixed probability that mutation will occur during the growth of the numerous independent cultures to saturation. Since the number of cell divisions, and hence the probability of mutation, can be fixed by limiting the available nutrient to any arbitrary level, a population size may be attained in which mutants will be found only in a certain percentage of the total independent cultures, enabling mutation rate to be calculated.

A third program has been initiated, in collaboration with Dr. Bo Prytz, on the genetic and chemical properties of bacterial mutants. This research is directed toward determining the biochemical properties and genetic stability of mutant strains of E. coli resistant to poisons and enzyme inhibitors. Dr. Prytz has been conducting a comparative quantitative analysis of the desoxyribose and ribose nucleic acid content of E. coli strains resistant to radiation and phages and dependent on streptomycin. The work with Dr. Prytz is being conducted under a government grant, and in this brief report a summary will be given of that

portion of our work which has been presented recently at a scientific meeting. Attention has been directed to a study of the response of various mutants of E. coli to radiation effects, extended to include the indirect influence of ultraviolet by way of irradiated media. It has been found that strains of cells resistant to irradiated media are those already known to be resistant to X-rays, ultraviolet radiation, and nitrogen mustard. A great quantity of ultraviolet radiation must be obtained from a mercury vapor lamp to demonstrate the indirect effect. A minimum of two hours' irradiation at 60 ergs per square millimeter per second will produce a maximum effect on cell viability. Further irradiation does not increase the proportion of inviable cells, but does influence the ability of surviving cells to multiply. For example, broth irradiated six hours will inactivate no more bacteria than another quantity of broth irradiated for only two hours. The percentage of inactivated cells is determined by the mutant strain. However, the ability of surviving bacteria to reproduce becomes progressively less as the prior period of exposure of broth to ultraviolet is lengthened. It appears possible at present that two effects of irradiation are being measured. One involves direct effects of products formed by the excitation of water—for example, hydrogen peroxide and hydroxyl radicals. Detoxification of irradiated broth by catalase proves that hydrogen peroxide is an important toxic constituent responsible for the initial rapid destruction or inactivation of cells. Peroxide is regarded as reaching a state of reversible equilibrium limiting the concentration. A second system involving oxidation of substrate materials in broth by peroxide would be cumulative with time and would eventually prevent surviving cells from growing. Even the cumulative secondary system is reversible, but a minimum of six hours following irradiation may be required before cell growth can occur. Evidence that oxidizing agents induced by irradiation are less toxic to radiation- and nitrogen-mustard-resistant strains of E. coli is being correlated directly with experiments with peroxides. Considerable support to the view that oxidizing agents are essential to the bactericidal or bacteriostatic effects of irradiated broth is given also by the finding that reducing agents such as ferrous ion, acting as electron donors, will neutralize the effect. Provision of sulfhydryl groups also will antagonize the inhibiting effect of irradiated broth. One of the indirect mechanisms of action of radiant energy may be oxidation of sulfhydryl groups in key enzyme systems to the disulfide state, thus effectively blocking protein synthesis. Study of the resistance of B/r and B/M, the radiation-resistant strains, to agents capable of alkylating or forming mercaptide bonds with SH groups has therefore been begun. If resistance to radiation is associated with a shift in the quantity or chemical bonding of SH groups, this should appear as a common property of radiation-resistant cells, apparent on exposure to a variety of SH-inactivating substances.

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Albert Kelner and Louise Pirovane

1. The Genetics of Antibiotic Formation

The studies reported in the Annual Reports of 1946 and 1947 were completed by October 1 of this year. The research on the genetics of antibiotic formation, from October 1946 until the present time, may be summarized as follows:

Seven actinomycete strains were selected from the American Type Culture Collection on the basis of their having either no, or only trace, antibotic activity against Micrococcus lysodeikticus, or Escherichia coli, or both of these bacteria. The actinomycetes were Streptomyces albosporeus ATC 3003, Streptomyces albus ATC 3004, Streptomyces cellulosae ATC 3313, Streptomyces flaveolus ATC 3319, Streptomyces griseus ATC 3326 (a non-streptomycin-producing strain), Streptomyces violaceus ATC 3355, and Streptomyces viridochromogenus ATC 3356. By means of irradiation with X-rays, or in some cases with ultraviolet light, as high a frequency of mutation as feasible was induced in conidial suspensions of these antibiotically inactive actinomycetes. Large populations of irradiated spores were then tested for the presence of mutants with antibiotic activity against a bacterium — either M. lysodeikticus, Staphylococcus aureus, or E. coli—against which the parent culture was inactive.

Antibiotically active mutants were found in all but two (S. albus and S. cellulosae) of the seven actinomycetes studied. Therefore it seems probable that most actinomycete strains, including those apparently inactive, can be induced to form antibiotically active mutants. A mutant originating from inactive S. griseus ATC 3326 produced an antibiotic that was either streptomycin or a closely related substance. Active mutants were present with a frequency ranging from 0.01 to 1.9 per cent. Many of the mutants were types that would probably not survive in natural habitats.

The mutants obtained from one parent culture differed from one another in antibacterial spectrum, suggesting that mutants forming diverse, qualitatively different antibiotics could be induced to form from a single parent culture.

Under the influence of such great pioneers in antibiotic research as Waksman and Fleming, this field has enormously advanced the welfare of the human race. If the science of antibiotics is to continue to advance, however, there is need for many more theoretical studies such as those described here. Otherwise the science will remain largely empirical, and its potentialities for further development will be greatly lessened.

2. Effect of Visible Light on the Recovery of Microorganisms from Ultraviolet-Irradiation Injury

Early in January, 1948, we became interested in the problem of the recovery of microorganisms after ultraviolet irradiation. Ultraviolet light injures most cells to such an extent that they are unable to grow or form a colony. We had observed that some of the cells of ultraviolet-irradiated

S. griseus spores recovered their ability to grow if stored for a day or two after ultraviolet irradiation. So little was known about the recovery phenomenon, and the implications of this phenomenon for genetics, medicine, and cellular physiology seemed so important to us, that we resolved to make an intensive study of the recovery of microorganisms from the effects of ultraviolet irradiation. In such time as we could spare from our main research on antibiotics, we initiated experiments designed to uncover the significant mechanisms causing this recovery. Toward the end of the summer, we discovered that illumination of ultraviolet-injured cells with visible light profoundly increased their recovery

After October 1, when our grant for antibiotic research terminated, we were very sorry to have to lose the invaluable help of our efficient assistant, Louise Pirovane. Subsequent research was done without the help of an assistant.

The kinetics of the reaction were determined, and the roles of ultraviolet dosage, visible-light intensity and duration, temperature coefficients, etc., were investigated, in order to put our discovery on a firm basis. Other laboratories to whom the findings were communicated confirmed the visible light-induced recovery or photoreactivation for other organisms than the actinomycetes, so that by the end of the year there was no doubt about the validity of the phenomenon.

The evidence suggested, then, that in visible light we have a factor which uniformly and reproducably causes the recovery of many of the cells that are rendered nonviable by ultraviolet irradiation. The action is probably directly on the cells rather than on the menstruum, and there is no evidence that any experimental artifacts are involved. The magnitude of the effects makes it likely that a key factor in the lethal action of ultraviolet light is influenced by visible light. Whether or not light-induced recovery bears a relation to other types of recovery previously recorded is difficult to say. All such studies, as well as studies on ultraviolet-induced mutation, must be re-evaluated on the basis of whether or not light-induced recovery played a part.

The powerful action of light in the resuscitation of ultraviolet-treated cells leads us to hope that further study of this phenomenon may yield clues to factors causing similar recovery from X-irradiation or irradiation from radioactive materials. There is thus the possibility of at least a partial physiotherapy of radiation injury.

3. Bibliography

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Abramson, Harold A., Cold Spring Harbor, N. Y.—Experiments were divided into two groups: those involving the study of the behavior of hydrogen peroxide in the skin, and those connected with the electrophoretic fractionation of pollens causing hay fever and asthma. Miss M. Loebl assisted in most of the experiments.—Attempts were made to introduce into the human skin, electrophoretically, oxidation-reduction indicators of various forms. Hydrogen peroxide was then transported electrophoretically into the same sites, with a view to studying the in vivo behavior of peroxides. These experiments were unsuccessful. A technique of studying the stability of glycerite of hydrogen peroxide on human skin was developed, however, by using starch iodide paper. It was found with this technique, contrary to expectation, that on the normal skin glycerite of hydrogen peroxide decomposes very slowly, so that peroxide is present for more than one hour. This extends the field of usefulness of the nontoxic peroxide as a skin antiseptic.—Giant ragweed pollen was successfully purified by electrophoretic fractionation so that quantities of Trifidin, suitable for therapeutic trials, were obtained. A simplified technique permitted the performance of the fractionation at room temperature.

Adams, Mark H., New York University College of Medicine, New York, N. Y.—In further studies on the role of calcium ion in the multiplication of phage T5 on strain B of E. coli, the rate of adsorption of virus to host cell was determined in chemically defined medium in the presence and absence of calcium ion. The medium contained glucose, ammonium chloride, phosphate buffer, and magnesium sulfate. In the absence of added calcium, the velocity constant for adsorption was 3×10^{11} cm.^{3}/min., whereas in the presence of 10^{-3} M calcium chloride, the velocity constant was 4×10^{-11} cm.^{3}/min. These constants are probably not significantly different, and certainly do not account for the fact that phage T5 grows well on E. coli in the calcium-containing medium and not at all in the calcium-free medium. The latent period of T5 in the calcium-containing medium was 55 to 60 minutes, but the addition of glutamic acid to the medium decreased the latent period to 45 minutes, presumably by improving the nutritional state of the host cell.—In the glutamic-acid-containing medium, with no added calcium, phage T5 was rapidly adsorbed to the host cell, but no living phage was liberated. Instead, the number of infectious centers decreased rapidly, indicating a loss of infectivity of the virus after adsorption to the host cell. This does not occur if calcium is present. Apparently calcium is necessary not only for multiplication but also for survival of phage T5 in the host cell.

Bernheimer, Alan W., New York University, College of Medicine, New York, N. Y.—Although viruses specific for a wide variety of plants

and animals are known, none has been described for the Protozoa. Should protozoan viruses be found, they would undoubtedly prove extremely useful in studying virus-host cell relationships. Attempts were made to find viruses for a number of free-living organisms—namely, five strains of ciliated protozoa (Tetrahymena gellei), and the flagellates Euglena proxima, Polytoma uvella, and Chilomonas paramecium. These microorganisms, cultivated in peptone solution free from bacteria and other protozoa, were employed throughout the study.—Repeated collections of pond water were made from more than a dozen stations in Nassau County. Sterile filtrates of the pond water, as well as of pond water "enrichment cultures," were inoculated from the stock protozoan cultures, and the ensuing growth of protozoa examined for gross evidence of viral activity, such as lysis, loss of motility, and so forth. In addition, "blind passage," involving serial transfer of culture filtrates, was carried out. The experiments did not, in any instance, indicate the presence of the viruses sought. In spite of these negative results, it is considered not unlikely that the viruses postulated actually do exist, and that the materials and techniques employed were not adequate to reveal their presence.

Cohen, Seymour S., The Children's Hospital of Philadelphia and University of Pennsylvania, Philadelphia, Pa.—(1) The qualitative analysis of sugar phosphates by paper chromatography after enzymic hydrolysis: In the course of enzyme studies on the possible origin of ribose via the oxidative degradation of phosphogluconic acid, it became necessary to develop methods for the characterization of intermediate sugar phosphates, the sugar content of which was of the order of amounts of 100 micrograms or less. Various sugar phosphates were hydrolysed with purified intestinal phosphates at pH 9, and the resulting hydrolysate was examined in the paper chromatogram by the method of Partridge. It was found that the following sugar phosphates previously isolated from natural sources contained only the sugar designated: glucose-1-phosphate, glucose-6-phosphate, fructose-6-phosphate, fructose 1, 6 diphosphate, ribose-3-phosphate, and ribose-5-phosphate.—It was found, in collaboration with Dr. D. B. McNair Scott, that the primary oxidation product of 6-phosphogluconic acid yielded a material which, after enzymatic hydrolysis, had the chromatographic and chemical behavior of 2-keto-gluconic acid. The first enzymatic oxidation product of 6-phosphogluconic acid is therefore presumptively considered to be 2-keto-6-phosphogluconic acid.—(2) The correlation of desoxyribose nucleic acid (DNA) and virus synthesis in E. coli infected by T4r: In collaboration with Dr. A. H. Doermann at the Carnegie Institution, it was found that cells multiply infected with T4r at 30° had a latent period of about 36 minutes, a rise period of about 15 minutes, and a burst size of about 105-115 T4r particles, and started synthesizing DNA at 10-12 minutes. The rate of synthesis increased somewhat until 15%-20% of the maximum amount had been formed, and then continued linearly until about 36-37 minutes.

3.5×10^{-16} grams of DNA was found to be synthetized for every virus particle produced. Intracellular virus appeared at about 17-18 minutes, increased slowly, and continued linearly until about 44 minutes. The last 80% of the DNA and virus curves were parallel, DNA preceding virus by about 4-6 minutes. Clearly, the initial laying down of DNA does not complete a virus particle.

Lieb, Margaret, Columbia University, New York, N. Y.—A preliminary investigation was made of the toxicity and mutagenic activity of several commercial antithistamine preparations, using Escherichia coli. In other studies, various spontaneously appearing back-mutants from biochemically deficient strains were isolated and compared to the parent strain as to phage-resistance patterns, ability to ferment certain sugars, and stability on synthetic medium. In several cases, back-mutation was accompanied by an appearance of sensitivity to certain phages, while all the cells of the parent strain apparently remained resistant.—As the result of tests on "complete" and "minimal" media, the bacteriophage T3 was found to require one or more cofactors for lysis of E coli strain K12; casamino acids showed little or no cofactor activity.—These studies are being continued at Columbia University.

Luria, S. E., and Dulbecco, R., Indiana University, Bloomington, Ind. —During the summer we continued the study of the effect of ultraviolet radiation on bacteriophages labeled with genetic markers. In particular, the problem being attacked at present is whether production of lethal mutations by radiation causes the elimination of the determinants of specific characters. The characters used were the pair of alternative characters r^+ and r22 and the pair h^+ and h, in bacteriophage T2. We also studied a different type of T2r mutant, indicated as r23, which is distinguishable phenotypically from r22 because the plaque formed by it is fuzzy instead of being sharply delimited as that of r22. We actually found that all the r mutants isolated from T2L are phenotypically either like r22 or like r23. The experiments were based on the principle of mixed infection with one particle of one active phage and one particle of phage carrying the alternative character and exposed to various radiation doses. For example, we used $T2r^+$ active and T2r22 irradiated. Single infection with both phages was obtained by using low multiplicities, so that only a few bacteria had one phage particle of both types. The bacteria having phage of both types would be expected to liberate a mixture of the two mutants—for example, r^+ and r—unless the corresponding locus had been inactivated by irradiation. The proportion of bacteria that liberate both r^+ and r phages was determined by the technique of mottled plaques, checked by numerous large-scale single-burst experiments. By this method, it was found that there was a logarithmic loss of the locus r22 when irradiated with increasing doses of ultraviolet light. The rate of loss of the locus r22 was approximately one-fourteenth of the rate of loss of phage activity of the phage particle

itself. When the reverse cross was made, however, namely, r22 active and $r(22)^+$ inactive, it was found that the rate of loss of the $r(22)^+$ locus was also logarithmic, but with a slope approximately twice as great as that of the r22 locus, as though the allele r^+ had a sensitivity twice as great as that of the allele r22. Experiments of the same type with the locus h gave results comparable to those with r22, with the complication that the h locus appeared to be eliminated in some of the bacteria containing also bacteriophage h^+, even in the absence of irradiation. The single-burst experiments, while confirming the validity of the mottled plaque method, supplied the additional information that the bacteria, which after having been infected with one active particle and one irradiated one liberate both phages $T2r^+$ and T2r22, yield fewer particles carrying the allele that comes from the irradiated particle. This shows that the incorporation into the final particles of loci coming from different infecting particles is not independent of the presence or absence of lethal mutations in the infecting particles, even if the lethal mutations do not directly affect the locus being started. Whether this is an expression of linkage or of some other phenomenon remains to be seen. As a result of this fact, the slopes of the inactivation curves for individual loci (see above) cannot be taken as indications of the rate of inactivation of the locus by ultraviolet radiation and may, at best, give an indication of the upper limit of the sensitivity of the locus. The orders of magnitude of the sensitivity found for the loci r22, $r,^+$, and h appear to be compatible with the estimate of 25 for the minimum number of independently transferable loci at which lethal mutations can be produced by ultraviolet radiation.—In the course of the summer a number of double mutants of bacteriophage T2 were isolated in order to study, through experiments of the type discussed above, the elimination by radiation of loci with various degrees of linkage as defined by Hershey.

Mayr, Ernst, American Museum of Natural History, New York, N.Y. —Work was continued on the interrelation between Yucca pollination and the Yucca moth. The results of last season were confirmed: apparently some Yucca flowers are being pollinated by agencies other than the Yucca moth.—The major portion of my time was devoted to writing several chapters of a book entitled "The Biology of Birds."

Menkin, Valy, Temple University School of Medicine, Philadelphia, Pa.—Severely injured cells of vertebrate material seem to release in inflammatory exudates a toxic material located in or associated with the euglobulin fraction of exudates. This toxic substance, which, per se, offers a reasonable explanation for the basic pattern of injury in inflammation, has been termed necrosin.—What appears to resemble this state of affairs is likewise encountered in severely damaged tissues of some invertebrates. Crushed tissues of either the soft-shell clam (Mya arenaria) or the gills of the horseshoe crab (Limulus polyphemus) yield an extract which is toxic to otherwise normal Mya arenaria. The toxicity seems to reside

primarily in the protein fraction obtained by treating the extract with $(NH_3)_2SO_4$ at one-third saturation. This chemical procedure in vertebrate material yields after dialysis of the sulfate ions what is termed the euglobulin fraction. It is in association with the euglobulin fraction of the inflammatory exudates that necrosin is recovered.—The toxic euglobulin factor in the injured tissue of the invertebrates studied is thermolabile. Boiling tends to inactivate it in large part. This is precisely the same property possessed by necrosin.—The studies indicate and suggest that the toxic factor in the severely injured tissues of the invertebrate forms studied is similar, if not identical, to necrosin recovered from the products of cellular injury in vertebrates.—There may be an additional thermostable toxic factor, to be studied further, in the injured tissues of Mya arenaria. This study is based on over 200 clams.

Michaelis, L., Granick, S., and Wollman, S. H., Rockefeller Institute for Medical Research, New York, N. Y.—Studies on the staining properties of fixed tissue sections were continued, especially with respect to acid dyes and some processes connected with the Feulgen reaction of nucleic acids.—The method of paper chromatography was applied for the separation of various porphyrin compounds. Preliminary experiments were aimed at finding the most suitable solvent. The method was tried both for animal iron porphyrin compounds and for vegetable magnesium porphyrin compounds (chlorophyll). Separation of closely related compounds has been successful and will be undertaken in detail in the near future.—Studies on some of Dr. McClintock's corn seedling mutants were undertaken, in the expectation of finding precursors of chlorophyll such as have been obtained by Dr. Granick in chlorella. None could be detected, however.—Studies of some brown and red algae were started with a view toward characterizing the so-called chlorophyll-C pigment.

Sheldon, W. H., College of Physicians and Surgeons, New York, N. Y.—The work I did at the Laboratory last summer consisted in a final revision and finishing of the book "Varieties of Delinquent Youth." Also we did some preliminary work on another monograph in Constitutional Medicine which will be published as an "Atlas for Somatotyping Men."—I should like to add that the privilege of being with you last summer was greatly appreciated and that the stay out there was pleasant indeed. The book "Varieties of Delinquent Youth" is scheduled for publication in June 1949.

Stent, Gunther S., California Institute of Technology, Pasadena, Calif.—The virus-antiserum reaction of bacteriophage deviates from first-order kinetics after the percentage of survivors has become small; the point at which this deviation becomes noticeable varies between different strains of phage. This behavior may mean either that the phage is initially heterogeneous with respect to ease of inactivation or that active phage becomes progressively more difficult to inactivate with prolonged exposure

to antiserum.—Using the second alternative as model, a kinetic equation was derived. It was assumed that adsorption of an antiserum molecule at a certain one of n possible sites on the phage is required for inactivation of the phage and that the rate at which antiserum molecules are adsorbed to any one of the n sites is proportional to the a + 1 power of the fraction of the n sites still remaining vacant (where a is some constant equal to or greater than zero). The usual assumption that the rate is inversely proportional to the dilution (D) of the antiserum was also made, and the integrated form of the kinetic equation follows: $\ln P = -(1/a) \ln(KatⁱnD + 1)$, where P is the fraction of phage surviving after time t and K the reaction rate constant.—The antiserum inactivation kinetics were measured accurately for strains T3 and T5; and, after appropriate values for a and K were chosen for these experiments and for one reported in the literature on T1, the theoretical curves predicted from the kinetic equation agreed with experimental data throughout the range of measurement. Thus the present quantitative formulation of the assumption of progressive resistance to inactivation is compatible with observation.—(2) Bacteriophage "inactivated" by certain means, for example ultraviolet light, still interferes with bacterial reproduction, presumably by still being capable of being adsorbed to the bacterium. It was therefore of interest to see whether T5 inactivated in the absence of divalent ions at 37°, or in the presence of divalent ions at 65°, or by antiserum action, still has the ability to kill bacteria. T5 phage, inactivated more than 99% by each of the above methods, was mixed with bacteria under the conditions usually employed for multiple infection studies, and the surviving bacteria assayed by colony counts. Controls with active T5 phage and addition of no phage were carried out simultaneously, and it was found that only in the case of active phage was the bacterial count considerably reduced. The counts for phage inactivated by the three methods were essentially the same as when no phage at all had been added. It may therefore be concluded that T5 inactivated in the ways described does not interfere with bacterial growth.

Watson, James D., Indiana University, Bloomington, Ind.—During the summer I investigated further the reactivation of bacteriophage that had been inactivated by X-rays. Bacteriophage samples were exposed to various doses of X-rays at Memorial Hospital in New York City. The present status of the problem is the following:—The X-ray-inactivated particles of T2 are still able to kill bacterial cells, although in reduced amount. The rate of suppression of ability to kill bacterial cells is a logarithmic function of the dose of radiation, with approximately one out of three inactivating hits suppressing the ability to kill. In the case of active phage particles or particles inactivated by ultraviolet light, killing always follows adsorption of at least one phage particle. The rate of bacterial killing in these cases can be used as a measurement of phage adsorption. At present, it appears likely that this relationship also holds for phage inactivated by X-rays, although it has not been proved. Preliminary

experiments suggest that the loss of killing ability following X-ray irradiation occurs also with T6. T2h and T2r22 also behave like wild-type T2.—The particles that are adsorbed by bacteria do give a certain amount of reactivation upon multiple infection, as indicated by the fact that the bacteria that yield active phage are more numerous than those that receive the residual active particles. The ability of X-rayed bacteriophage T2 to give genetic recombination and reactivation was confirmed by experiments with the mutant T2r22. An inactive particle of phage T2r can contribute the r locus and give mixed yields if crossed with $T2r^+$ either active or irradiated with ultraviolet light. Further proof that X-ray-inactivated bacteriophage could actually be reactivated was obtained in experiments in which X-ray-inactivated phage T2r was reactivated upon mixed infection with bacteriophage $T6r^+$. It was also found that reactivation of X-ray-inactivated bacteriophage occurs with phage T6 as well as with T2. Phage T6 behaves almost exactly as phage T2, in so far as the probability of reactivation is concerned.—The probability of reactivation and of genetic transfer involving the X-ray-inactivated bacteriophages is much lower (by a factor 2-3) than that corresponding to bacteriophage particles inactivated by ultraviolet light with the same number of inactivating hits. It is difficult at the present stage of these experiments to decide on an interpretation of this fact. It is possible that each act of adsorption of X-rays either affects several genetic determinants or reduces the probability of successful transfer of active loci from an inactive particle to another. Unlikely, though not yet excluded, is the possibility that X-rays may reduce the probability of reactivation because they also produce in the bacteriophage other genetic changes that cannot be restored by genetic transfer.

COURSE ON BACTERIOPHAGES

June 28-July 17, 1948

Instructor: Mark H. Adams, New York University, College of Medicine.

Assistant: Nancy J. Collins, New York University.

For the fourth successive year, an intensive three-week course was offered dealing with techniques and problems of research on bacterial viruses. Fourteen students were formally enrolled in the course, and two more attended the discussions and seminars without doing the laboratory work. Two new experiments were added to the course schedule, one involving a study of the kinetics of interaction between viruses and their antibodies, the other dealing with the effects of ultraviolet light on viruses. After the conclusion of the course several students undertook research work on bacterial viruses. The virus course and subsequent virus research work were housed in the Davenport Laboratory.

The following students were enrolled in the course: Dr. S. Benzer, Department of Physics, Purdue University; Mr. Ernst H. Beutner, Department of Bacteriology, University of Pennsylvania; Dr. B. D. Davis, U. S. Public Health Service, and Cornell Medical School, New York, N. Y.; Dr. Joseph S. Gots, Department of Bacteriology, University of Pennsylvania School of Medicine; Mr. Felix Haas, Department of Zoology, University of Texas; Miss Lillian Jedeikin, Children's Hospital, Philadelphia, Pennsylvania; Miss Margaret Lieb, Department of Zoology, Barnard College; Miss Ethelyn Lively, Carnegie Institution of Washington, Cold Spring Harbor, N. Y.; Dr. Ilda McVeigh, New York Botanical Garden, New York, N. Y.; Dr. Harold T. Meryman, Naval Medical Research Institute, Bethesda, Maryland; Dr. Morris Schaeffer, City Hospital, Cleveland, Ohio; Dr. Gunther Stent, Department of Chemistry, University of Illinois; Mr. Dudley Thomas, Department of Chemistry, California Institute of Technology; Dr. Seymour Wollman, Rockefeller Institute for Medical Research, New York, N. Y.

It is interesting to note that, of the 47 individuals who have taken the bacteriophage course during the past four years, 30 have been active research workers with a doctorate, and most of the remainder graduate students working towards an advanced degree.

In connection with the phage course a series of seminars, attended by students in the course and anyone else interested, was given by research workers at Cold Spring Harbor and by visitors. The speakers and topics are listed below:

S. E. Luria—Reactivation of ultraviolet-inactivated phage.

R. Dulbecco—Application of statistical methods to phage research.

A. H. Doermann—Release of intracellular phage during the latent period.

H. T. Meryman—The use of the electron microscope.

E. Witkin—Effect of chemicals on bacterial mutation rates.

F.Haas—Increase of bacterial mutation rates by ultraviolet irradiation of the growth medium.

R. Foster—Effect of acridines on phage-infected E. coli.

R. Goldwasser—Phosphorus metabolism in phage-infected Staphylococci.

F. Putnam—Purification and properties of T6 phage and phosphorous metabolism of infected E. coli.

B. D. Davis—Principles of chemotherapy.

NATURE STUDY COURSE

J. Southgate Y. Hoyt, Ph.D

Sally F. Hoyt, Ph.D.

Cornell University, Department of Conservation, Laboratory of Ornithology, Ithaca, N. Y.

Nature study at the Cold Spring Harbor Biological Laboratory, now in its fifth year, has established an enviable reputation both with the students participating and with those administrating the course. It is not often that one has the opportunity to see the results of one's work accepted as enthusiastically and with such willing participation from all concerned as has been shown in the work with the children in the Nature Study courses. The enrollment has grown to the maximum each year, and another student would have taxed the class.

As in the past, the course was conducted with four aims in view: (1) that the students learn to observe the many interesting plants and animals that occur abundantly about them all the time; (2) that each student learn how to make observations of natural phenomena and how to answer his own questions about his findings; (3) that they learn how to make, study, and care for collections, as well as the purposes of having collections; (4) that they learn that each person can make a contribution by careful and accurate study of the smaller incidents observed by all of us but not interpreted and heeded, in the tremendous field for study always at hand.

The Biological Laboratory nestles in an unusually rich location for these studies. Within short walking distance one may visit large fresh-water ponds; swift, heavily shaded streams; hot, slow streams; the merg-ence of fresh-water streams into salty lagoons, providing brackish water habitats; heavy forest areas of deep shade and moist floor; open fields, with typical field flora and fauna; long, sandy beach and tide pools among rocks; and rich, deep salt swamp and mud flats. Each of these places was visited by some class, and the life found was studied with emphasis on how much could be seen and its relation to other forms of life in nearby areas. Wawepex Laboratory served as headquarters for the course; it was here that the students gathered for more detailed study of the forms found in the field. The laboratory was open at all times, and each student was encouraged to return at any time for any type of work he wanted.

There were three age groups, divided as follows: the Juniors, aged six to eight, met every Monday and Wednesday from 9:00 to 11:00 a. m., and consisted of twenty-two students; the Intermediate group, aged nine to eleven, met every Tuesday and Thursday from 9:00 to 11:00 a. m., and consisted of eighteen students; the Advanced group, aged twelve and older, met every Monday and Wednesday from 2:00 to 4:00 p. m., and consisted of ten students. There was also an adult class of five students, who requested that a class be formed for them. More will be said about

this class later; it met every Monday and Wednesday evening from 7:00 to 9:00.

The members of the youngest group, the Juniors, spent a large part of their time roaming the various regions in the vicinity of the laboratory in search of everything they could find, learning that in every corner there is some form of life especially adapted and suited for its special existence in that particular habitat. Two questions are always asked first of all: (1) "What is it?" and (2) "What will it do, or what good is it?" Whenever possible the students found the answers to these questions themselves and learned to relate one form of life to some other seen before, either in the class or elsewhere. On one field trip, a member of the class spotted an enormous bright-green caterpillar hanging on a sassafras tree. The class soon learned the value of camouflage in nature and started looking for more examples. Starting with this caterpillar, they became very much interested in the life of butterflies and moths and started a collection of such animals.

With the help of the class we found several Monarch Butterfly caterpillars and chrysalids to watch. Unfortunately, no member of this class saw the actual transformation from one stage to another, but they all saw the results and learned about the life history of many insects in this manner.

The Intermediates did much the same things as the Juniors, but a little more strenuously and with a slightly more serious and fundamental approach. This group conducted an interesting experiment with larvae of caddis flies collected in the stream of the fish hatchery. Having removed the larvae from their natural homes or casts, the class provided them with all types of material for constructing a new cast, and watched to determine what preference, if any, they showed for the various materials supplied. Some artistic casts were produced by providing the larvae with brightly colored pieces of flower petals. This was very interesting; it formed the basis of a long lesson on the experimental method of observation, and served to set a high standard for future work done by this class.

This group visited a pond on the estate of one of the members of the class, and with nets they produced findings that astounded even the instructors. Bullfrog tadpoles measuring six inches in length were found by the scores, as well as hundreds of silver carp and sunfish. These were studied with great interest and produced material for many long discussions on habitat, food habits, and requirements for life. Another similar-looking pond on the same estate was examined and found to be almost devoid of such life as was found in the first pond. This again afforded a wonderful opportunity for comparative study of the requirements for these forms of life.

The Advanced group was small, and was united in an interest in

the unknown life of the waters around the community. Their interest seemed to concentrate on the microorganisms for quite a long time, and consequently we spent a great deal of time collecting material from all possible sources and studying it in the laboratory. Without exception, every member of the class was excited and anxious to learn the names and habits of as many of the microorganisms as possible. A large number was seen by the class, including a few rather uncommon forms. During the latter part of the course this group became interested in studying and collecting moths, butterflies, and other of the larger, more spectacular insects. A trip was taken by car to some fields and woods not visited by other classes, and here new material was collected with the aid of home-made as well as laboratory-provided nets. This material was mounted, and each student took home with him some attractive mounts and beautiful collections of insects. Sharing of experiences and exchange of methods was especially encouraged among the members of a class, with the result that all learned from others how to do something not known before.

All the classes were taken to the New York State Fish Hatchery, across the highway from the Biological Laboratory. Mr. Walters, the director, very kindly provided an escort for the classes, so that we had ample time to study and observe the methods employed and the reasons for conducting such work. The classes showed keen interest in the demonstrations of the development of trout from eggs to adults in the tanks. Special emphasis was placed on the role the hatchery plays in the conservation of our resources.

The Roosevelt Bird Sanctuary in Oyster Bay was also visited by all the classes. Here Mr. Callaghan, the director, very generously explained all the exhibits and purposes of the sanctuary. The classes were able to see some forms of life we had been unable to study at the laboratory in the short time allotted to each class. The excellent demonstrations displayed in the museum of this sanctuary were especially appreciated and studied very carefully by many of the students.

Each class visited the sand spit both at high tide and at low tide, to observe the differences in life at the two times. Some very interesting collections were made and brought back to the laboratory for further study.

All the students were encouraged to bring from home anything they could find of interest for the rest of the class to see. Many students caught quite a few insects, including many of the more spectacular larger moths. They were shown how to prepare these for mounts, and some even prepared mounts of a permanent nature for display in their homes. A few students were interested in starting collections of pressed flowers. Help was given here, and suggestions were offered as to methods of collecting and pressing and keeping the flowers, but the students were encouraged to do most of the work for themselves. One rainy day was spent

in making leaf prints from leaves collected and pressed during a previous walk. One class had an opportunity to see how bird-banding is carried on, when a student brought in a thrush which he had raised from a nestling, and later released it in time for it to migrate. A girl was especially interested in finding as many different shells as she could during the period of the course. It was found that she had collected some twenty different kinds by the end of the course. These were displayed, and she was helped in learning the name of each one and the reasons for the various shapes as well as the lives of the animals that had once inhabited them. Similar instances can be mentioned for almost any field of natural history one may choose.

The adult class was formed by request for lectures on the various fields of natural history. An hour-and-a-half lecture was followed by a half hour of demonstration and discussion. The subjects covered were lower plants, flowering plants, invertebrates, insects, fish amphibia, reptiles, birds, and mammals. About two periods were devoted to a subject and each lecture attempted to give a working background in the field so that a person would know how to use the various references in learning more about the subject. Several early-morning field trips were taken—principally for the study of birds, but anything that presented itself was observed and discussed. If time permitted, demonstrations were made of the techniques employed in preparing material for collections, but this phase was cut short for lack of time in every case.

August 5 was the formal closing of the course, with a public exhibition of the activities of the various classes. The students were encouraged to prepare demonstrations of their particular interests and to arrange the material in an attractive manner for inspection by their parents and friends. Without exception, every member of the course had some type of exhibit prepared by himself, and many had several exhibits. At two o'clock the parents and friends assembled in Wawepex Laboratory for the exhibition. Everyone was greatly impressed with the tremendous amount of material found within the few weeks of the course and gathered from this one locality. At three o'clock everyone was invited to attend an illustrated talk by Dr. Hoyt. This lecture presented an imaginary field trip of the classes, showing all the interesting and amazing findings that each student encounters as he goes along the paths through the woods, along the beaches, and across the fields. This pleasant field trip ended with ice cream and cake served to all present by the Laboratory and with a very informal social gathering of all parents and students.

Dr. and Mrs. Hoyt were assisted this year by a voluntary assistant, Miss Elizabeth Hawkins, who helped in the general conduct of the classes and in the care of the materials collected. Miss Hawkins intends to take up Biology in college and was anxious to have the opportunity of working with classes in this field.

The following is a list of students who attended the classes:

Abramson, Barbara
Abramson, Harold, Jr.
Ames, Edward
Ayer, Douglass
Ayer, William
Baikow, David
Baty, Larilee
Bryson, Constance
Buckley, Christine
Cleaveland, Edward
Cox, Basil
Dobzhansky, Sophie
Frenkel, John
Funnell, Eleanor
Gardner, Wendy
Gillmore, Carol
Gillmore, Lynn
Grace, Catha
Granick, Donna
Hawkins, Ashton
Hestwood, Deborah
Kaufmann, Anders
Lefferts, Peter
Little, Billy
Lomasney, Lynn
Martin, Peter
Martin, William
Mayr, Susie
Menkin, Gabriel
Menkin, Lucy
Muhlhausen, Peter
Mutter, Frederick
Mutter, Valerie
Myer, Betty
Nields, Elizabeth
O'Connell, Raymond
Olds, Pepper
Olmsted, Nancy
Pierce, Martin
Rutherford, John
Schwartz, Arthur
Seaman, Robert
Sheshunoff, Alex
Truslow, Betsy
Truslow, Fred
Vacquier, Victor
Warner, Bradford, Jr.
Warner, Miner Hill
Weber, Derek
Weber, Tommy

COLD SPRING HARBOR SYMPOSIA ON QUANTITATIVE BIOLOGY

Current volume: XIII. Biological Applications of Tracer Elements. 220 + xii quarto pages, 87 figures. 1948. Table of contents is listed below:

Gurin, Samuel, and Crandall, Dana I.—The biological oxidation of fatty acids.

Hevesy, G.—Historical sketch of the biological application of tracer elements.

Kamen, Martin D., and Spiegelman, S.—Studies on the phosphate metabolism of some unicellular organisms.

Norris, William P., and Kisieleski, Walter—Comparative metabolism of radium, strontium, and calcium.

Rittenberg, D.—The application of the isotope technique to the study of the metabolism of glycine.

Sacks, Jacob—Mechanism of phosphate transfer across cell membranes.

Shemin, David—The biosynthesis of porphyrins.

Ussing, Hans H.—The use of tracers in the study of active ion tranport across animal membranes.

Wood, Harland G.—The synthesis of liver glycogen in the rat as an indicator of intermediary metabolism.

Previous Volumes

* Vol. I (1933) Surface Phenomena, 239 pp.
* Vol. II (1934) Growth, 284 pp.
* Vol. III (1935) Photochemical Reactions, 359 pp.
* Vol. IV (1936) Excitations, 376 pp.
* Vol. V (1937) Internal Secretions, 433 pp.
* Vol. VI (1938) Protein Chemistry, 395 pp.
* Vol. VII (1939) Biological Oxidations, 463 pp.

Vol. VIII (1940) Permeability and the Nature of Cell Membranes, 285 pp.

* Vol. IX (1941) Genes and Chromosomes, 315 pp.

Vol. X (1942) The Relation of Hormones to Development, 160 pp.

Vol. XI (1946) Heredity and Variation in Microorganisms, 314 pp.

Vol. XII (1947) Nucleic Acids and Nucleoproteins, 279 pp.

* Out of print.

LABORATORY STAFF

* Bass, Emma—Maid

Bryson, Vernon—Research Biologist

Demerec, M.—Director

Doermann, Harriet—Research Assistant

Dorsey, Henry—Laborer

Farrington, Margaret—Technical Assistant

Franzese, Eleanor—Clerical Assistant

* Hoyt, J. Southgate Y.—Nature Study Instructor

* Hoyt, Sally F.—Nature Study Instructor

* Kaufmann, Berwind N.—Stockroom man

Kelner, Albert—Bacteriologist

Klem, Dorothy—Secretary

Mann, Margaret—Research Assistant

Pirovane, Louise—Research Assistant

Prytz, Bo—Chemist

Rae, William S.—Superintendent of Grounds

Reddy, William—Laborer

Swanstrom, Maryda—Research Assistant

* Thompson, Stella—Dining Hall Manager

Yongen, Eileen—Research Assistant

* Summer

SUMMER RESEARCH INVESTIGATORS

Abramson, Harold A.—Cold Spring Harbor, N. Y.

Adams, Mark H.—New York University College of Medicine, New York, N. Y.

Bernheimer, Alan—New York University College of Medicine, New York, N. Y.

Cohen, Seymour S.—Children's Hospital, Philadelphia, Pa.

Collins, Nancy—New York University College of Medicine, New York, N. Y.

Delbruck, M.—California Institute of Technology, Pasadena, Calif.

Dulbecco, R.—Indiana University, Bloomington, Ind.

Ehrensvard, Gosta—University of Stockholm, Stockholm, Sweden.

Granick, S.—Rockefeller Institute for Medical Research, New York, N. Y.

Jedeikin, Lillian—Children's Hospital, Philadelphia, Pa.

Lieb, Margaret—Columbia University, New York, N. Y.

Loebl, Margot—New York, N. Y.

Luria, S. E.—Indiana University, Bloomington, Ind.

Mayr, Ernst—American Museum of Natural History, New York, N. Y.

Menkin, Valy—Temple University School of Medicine, Philadelphia, Pa.

Michaelis, L.—Rockefeller Institute for Medical Research, New York, N. Y.

Pearlman, Gertrude—Rockefeller Institute for Medical Reasearch, New York, N. Y.

Rader, Dora—New York University, New York, N. Y.

Reiner, John—Tufts College Medical School, Boston, Mass.

Saifer, A.—Veterans Hospital, Brooklyn, N. Y.

Sheldon, W. H.—College of Physicians and Surgeons, New York, N. Y.

Shemin, David—College of Physicians and Surgeons, New York, N. Y.

Siegel, Richard—Indiana University, Bloomington, Ind.

Stent, Gunther—California Institute of Technology, Pasadena, Calif.

Ussing, Hans H.—University of Copenhagen, Copenhagen, Denmark.

Waelsch, H. B.—New York Psychiatric Institute, New York, N. Y.

Waelsch, Salome—Columbia University, New York, N. Y.

Watson, James—Indiana University, Bloomington, Ind.

Wollman, Seymour—Rockefeller Institute for Medical Research, New York, N. Y.

REPORT OF THE SECRETARY

The 55th meeting of the Board of Directors of the Association was held on January 30, 1948 at the Down Town Association in New York City, with Vice-President Page presiding because of President Murphy's absence in New Zealand. The following officers of the Association were re-elected: President, Robert Cushman Murphy; Vice-President, Arthur W. Page; Treasurer, Grinnell Morris; Secretary, E. Carleton MacDowell; and Assistant Secretary, Berwind P. Kaufmann. M. Demerec was re-elected Director of the Laboratory. The following members were appointed as the Women's Committee for 1948: Mrs. George S. Franklin, Mrs. Van Santvoord Merle-Smith, Mrs. Alvin Devereux, Mrs. George Nichols, and Mrs. Percy H. Jennings. One hundred and sixty-nine contributors to the Association in 1947 were elected Sustaining Members. The Secretary reviewed the minutes of the last meetings of the Board and the Executive Committee, which were approved. The Laboratory Director then gave his report on current research, progress of the latest volume of the Symposia, and plans for the 1948 symposium and other summer activities. He also spoke of the Laboratory's association with the local Hospital, through consulting service and joint research projects. The Treasurer, in giving the financial report, recommended retaining an auditing firm, reorganizing the bookkeeping methods, and ending the fiscal year of the Association in the spring rather than in December. The Treasurer's report was unanimously approved, and the retention of Main and Company as auditors was authorized. The Laboratory Director distributed and discussed the proposed budget for 1948, and gave an analysis of the sources of income of the Laboratory. He recommended provision in the budget for the salary of Dr. V. Bryson, as research biologist. The proposed budget was unanimously accepted. A committee, consisting of Messrs. Morris, Nichols, and Redmond, was appointed to consider the question of Dr. Fricke's property in connection with the larger problem of the use of the Townsend Jones property as a contribution to the advancement of science.

The 25th Annual Meeting of the Association was held on September 9, 1948 at Blackford Hall. After the meeting had been opened by Dr. Murphy, the Secretary reported the chief acts of the Association since the last Annual meeting. Dr. Demerec presented his report of the activities of the Laboratory, including the research program, Symposium, course on bacteriophage, Nature Study Course, and work of summer visitors. He also reported repairs and improvements to the physical plant, mentioned the need for a new lecture hall, and renewed the invitation to members of the Association and their friends to visit the laboratories. The Treasurer's report was then distributed and discussed by Mr. Morris. New features of the financial statement included an audit by a firm of certified public accountants, change of the end of the fiscal year from December 31 to April 30, a policy of allotting specific securities to the trust funds

granted for specific purposes, and inclusion of the contributions received through the Women's Committee with other contributions. The Treasurer pointed out the deficit shown by the financial report, and emphasized the serious need for increased contributions. A nominating committee, consisting of Mrs. H. A. Abramson, Dr. Mark Adams, and Dr. E. Caspari, proposed the following for re-election to the Board of Directors, in the Class of 1952: W. H. Cole, Mrs. George S. Franklin, E. C. MacDowell, William B. Nichols, Roland L. Redmond, and B. H. Willier. These nominees were elected by unanimous vote.

The 56th meeting of the Board of Directors was deferred from July until September 9, at which time a motion of adjournment was voted and the meeting was finally held on September 30, 1948, at the Down Town Association in New York City. The minutes of the January meeting of the Board were reviewed by the Secretary. The Executive Committee was re-elected for the ensuing year, as follows: Robert Cushman Murphy, President; Arthur W. Page, Vice-President; Grinnell Morris, Treasurer; E. Carleton MacDowell, Secretary; Mrs. George S. Franklin, Chairman of the Women's Committee; William B. Nichols; and John K. Roosevelt. Mr. Morris discussed the recommendation of the auditors with regard to allocation of security investments to the endowment and special funds of the Association; and by unanimous vote the proposed allocations and additional purchases indicated by the Treasurer were approved. Dr. Demerec reported on the financial situation of the Laboratory, in terms of estimated expenses per year, estimated income, and amount required from dues and contributions to meet the difference between these items. The President appointed Mr. Nichols chairman of a Committee on Public Relations, with power to select its other members. Mr. Morris reported, for the special Committee on Sale of Property, that a tentative agreement with Dr. Fricke had been reached in regard to the completion of sale of land. The resolutions and legal form of indenture presented in connection with this agreement were unanimously adopted and approved. The President was authorized by vote to sign the contract between the Association and the United States Government for genetical and biochemical research at the Laboratory during the year beginning October 1, 1948.

E. Carleton MacDowell, Secretary

MAIN AND COMPANY

Certified Public Accountants

New York, U. S. A.

AUDIT CERTIFICATE

Long Island Biological Association,
Cold Spring Harbor, L. I., N. Y.

We have made an examination of the accounts of the Long Island Biological Association for the year ended April 30, 1949. Our examination was made in accordance with generally accepted auditing standards, and accordingly included such tests of the accounting records and such other auditing procedures as we considered necessary in the circumstances.

In our opinion, the accompanying balance sheet and statements of income and expense, net worth, and supporting schedules, together with the notes thereon, present fairly the position of the Long Island Biological Association at April 30, 1949 and the results of its operations for the year ended on that date.

Main and Company
Certified Public Accountants.

New York, N. Y.,
June 15, 1949.

LONG ISLAND BIOLOGICAL ASSOCIATION

BALANCE SHEET

APRIL 30, 1949

ASSETS

General and Endowment Fund			
Cash:			
In banks	$ 9,560.09		
On hand	50.00	$ 9,610.09	
Investments (market value $32,541.30)		31,656.87	
Accounts receivable:			
United States Atomic Energy Commission	$ 8,099.59		
United States Department of the Army	6,339.24		
Miscellaneous	914.34	15,353.17	
Land, buildings and equipment, at cost or appraisal values:			
Land	$ 86,466.52		
Improvements to land	2,898.01		
Buildings	101,265.00		
Land and buildings leased from Wawepex Society	49,700.00		
Equipment	57,940.32	298,269.85	$354,889.98
Special Funds			
Due from General and Endowment Fund		$ 267.12	
Investments (market value $15,131.25)		15,710.00	15,977.12
Total			$370,867.10

LIABILITIES AND NET WORTH

General and Endowment Fund

Liabilities:			
Accounts payable		$ 15,476.93	
Deferred income		193.50	
Due to Special Funds		267.12	
Special grants:			
Josiah Macy, Jr. Foundation	$ 839.44		
The Jane Coffin Childs Memorial Fund for Medical Research	604.76		
National Tuberculosis Association	199.06	1,643.26	
Endowment Fund:			
Dr. William J. Matheson Bequest		20,000.00	
Net Worth		$317,309.17	$354,889.98

Special Funds

Blackford Memorial Fund:			
Principal		$ 5,000.00	
Charles Benedict Davenport Memorial Fund:			
Principal	$4,934.75		
Unexpended income	167.77	5,102.50	
Charles Benedict Davenport, Junior, Fund:			
Principal		1,037.12	
Temple Prime Scholarship Fund:			
Principal	$2,500.00		
Unexpended income	62.50	2,562.50	
Dorothy Frances Rice Fund:			
Principal	$2,229.16		
Unexpended income	45.84	2,275.00	15,977.12
Total			$370,867.10

NOTE: In accordance with the Association's established practice, the above balance sheet does not include the inventory at April 30, 1949 of the published volumes of the Association's yearly Symposia of Quantitative Biology, nor has any depreciation or amortization on public buildings and equipment been recorded on the Association's records.

LAND, BUILDINGS AND EQUIPMENT

April 30, 1949

Land:			
Purchased with funds raised through public subscription		$69,466.52	
Henry W. de Forest land		12,000.00	
Airslie land		5,000.00	$ 86,466.52
Improvements to land:			
Pipe line		$ 1,860.39	
Road		746.64	
Light and telephone poles		290.98	2,898.01
Buildings:			
Airslie building		$ 5,000.00	
Blackford Hall *		19,000.00	
Cole Cottage		2,105.00	
Davenport Laboratory		8,500.00	
Henry W. de Forest Building		15,000.00	
Reginald G. Harris House		8,500.00	
Dr. Walter B. James Laboratory		13,500.00	
George L. Nichols Memorial Laboratory		13,700.00	
Williams House		11,300.00	
Urey Cottage		2,660.00	
Machine shop and garage		2,000.00	101,265.00
Land and buildings leased from Wawepex Society under lease expiring in 1979:			
Land		$13,500.00	
Buildings:			
Hooper House	$13,200.00		
Jones Laboratory	10,000.00		
Osterhout Cottage	5,500.00		
Wawepex Laboratory	7,500.00	36,200.00	49,700.00
Equipment:			
General		$38,577.27	
Biophysics		16,849.90	
Physiology		2,513.15	57,940.32
Total			$298,269.85

* Built on land leased from Wawepex Society

STATEMENT OF NET WORTH

For the Year Ended April 30, 1949

Balance, May 1, 1948		$317,461.22
Add:		
Excess of income over expense for the year ended April 30, 1949		105.07
		$317,566.29
Deduct:		
Transfer to principal of special funds representing prior years' additions out of income as follows:		
Dorothy Frances Rice Fund	$220.00	
Charles Benedict Davenport, Junior, Fund	37.12	257.12
Balance, April 30, 1949		$317,309.17

STATEMENT OF INCOME AND EXPENSE

For the Year Ended April 30, 1949

Income:			
Contributions:			
Dues and contributions	$ 6,232.09		
Carnegie Corporation (grant for annual Symposia)	6,000.00		
Wawepex Society	1,450.00	13,682.09	
Symposia:			
Book sales	$10,208.60		
Registration fees	62.55	10,271.15	
Dining hall		9,923.23	
Rooms and apartments		7,820.55	
Research fees		2,892.05	
Interest and dividends on investments		970.89	
Other income:			
Summer course tuition	$ 510.00		
John D. Jones Scholarship	250.00		
Nature study course	213.17		
Beach permits	144.56		
Annual distribution from Walter B. James Fund	125.00	1,242.73	
Total income			$46,802.69

Expense.

Symposia:			
Publication of annual Symposia on Quantitative Biology	$ 8,995.76		
Expense of participants and lecturers	3,144.25	$12,140.01	
Dining hall		9,247.55	
Rooms and apartments		1,776.54	
Research expenses		2,752.43	
Summer course expense		307.84	
Loss on sale of securities		155.87	
Buildings and grounds maintenance:			
Salaries	$ 6,758.36		
Materials and supplies	3,616.45		
Heat, light and water	2,078.60	12,453.41	
General and administrative:			
Salaries	$ 4,307.66		
Insurance	1,283.98		
Printing and stationery	568.42		
Telephone and Telegraph	430.15		
Miscellaneous	1,273.76	7,863.97	
Total expense			46,697.62
Excess of income over expense			$ 105.07

STATEMENT OF SPECIAL GRANTS

For the Year Ended April 30, 1949

FROM WHOM RECEIVED	Balance of Grant Unexpended 5/1/48	Transactions May 1, 1948 to April 30, 1949: Amounts Received	Expenditures Charged Against Grant	Income to Association Charged Against Grant	Balance, April 30, 1949: Due to Association (Accounts Receivable)	Unexpended Balance of Grant
Schenley Laboratories, Inc.	$7,304.04		$ 6,879.17	$ 424.87		
Josiah Macy, Jr. Foundation	1,624.89		785.45			$ 839.44
The Jane Coffin Childs Memorial Fund for Medical Research	303.17	$4,000.00	3,698.41			604.76
Brooklyn Cancer Committee		2,550.00	2,550.00			
National Tuberculosis Association		3,538.93	3,339.87			199.06
United States Atomic Energy Commission			8,099.59		$ 8,099.59	
United States Department of the Army		5,423.45	10,970.51	792.18	6,339.24	
Totals	$9,232.10	$15,512.38	$36,323.00	$1,217.05	$14,438.83	$1,643.26