LONG ISLAND BIOLOGICAL ASSOCIATION

ANNUAL REPORT

OF

THE BIOLOGICAL LABORATORY

COLD SPRING HARBOR
LONG ISLAND, NEW YORK
1950

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REPORT OF THE DIRECTOR

The year 1950 opened with very bright prospects for our Laboratory. In March the Carnegie Corporation of New York made a grant of $100,000 to the Carnegie Institution for the construction of an auditorium at Cold Spring Harbor, to be used jointly by the Biological Laboratory and the Department of Genetics. Later on, the Association received from the Rockefeller Foundation a grant of $45,000 for modernization of the Laboratory's present facilities.

The Carnegie Corporation grant should have answered the urgent need for more comfortable quarters for Symposia, conferences, and evening lectures, as well as indoor accommodations for recreational activities; and the Rockefeller Foundation grant should have provided ample funds for modernizing our dining room and residences and improving our grounds. Unfortunately for us, however, the Korean conflict arose, and created conditions that have had a tremendous effect on our planning. The auditorium was to have been erected as part of a larger building program involving laboratories for the Department of Genetics. Since the start of the Korean incident, building costs have increased about fifty per cent, and for a time it seemed doubtful that we would be able to go through with our building plans. Just as this report is written, however, the Executive Committee of the Carnegie Institution has approved plans for the construction of the laboratories; and it now seems probable that ground will be broken for the new buildings in the very near future. The Rockefeller grant, also, will not go so far under present circumstances. We will have to raise an additional fund of about $20,000 if we wish to complete the job of improvement and put the physical facilities of the Laboratory into good condition.

Again during the past year the Laboratory lost several good friends, with the passing of Messrs. Henry L. Stimson, George Nichols, and Harvey D. Gibson. Henry L. Stimson joined the Association in 1925, the second year of its existence, as a Patron. During the critical period of its organization he served on the Board of Directors (1925-1929) and gave much valued counsel. He resigned from the Board because his time was fully occupied by his important duties with the Federal Government. When the class of Members Emeriti of the Board of Directors was established, in 1943, he was the first to be elected. Mr. George Nichols was for many years a member and Patron of the Association; and, as a close neighbor, he was interested in the Laboratory and participated in many of its activities. Mr. Harvey D. Gibson was one of the group of members who have joined the Association more recently (1946) and taken an interest in the present affairs of the Laboratory.

Research

Our year-around research program continued during 1950 to produce interesting results in the two projects already under way. One of these is

concerned with hereditary changes in bacteria, particularly those responsible for the origin of strains resistant to streptomycin and other antibiotics. Dr. Bryson continued in charge of these studies, working in collaboration with Drs. B. Prytz and J. Hsie and with the help of four research and technical assistants. The funds to support this work came from the Army, the National Tuberculosis Association, and the Jane Coffin Childs Memorial Fund for Medical Research. In the second project, Dr. B. Wallace, with Dr. J. C. King and six assistants, carried forward the study of genetic effects produced by continuous exposure to ionizing radiation in populations of the fruit fly (Drosophila). This program was supported entirely by the Atomic Energy Commission.

Bryson Prytz, and their collaborators continued with analyses of the morphological and biochemical properties of strains of bacteria selected from a standard laboratory strain because of the possession of higher resistance to the effects of ultraviolet rays, bacterial viruses, or streptomycin or other chemicals. The recent studies of the group show that these mutations have definite effects on selective value in the presence of deleterious agents quite different from those against which they afford primary protection. This is another way of saying that most mutations have manifold effects, and that few can be regarded as neutral in the selective or evolutionary sense. The production of mutations in bacterial strains affords a method of changing the selective values, and hence the relative viabilities, of two or more types competing in the same environment. Emphasis is placed on the fact that a mutation need have no apriori relationship to the selective agent, governing a property not assumed to be of significance within the experimental environment. For example, the "lactose-negative" mutation in E. coli reduces viability in the presence of dinitrophenol in a broth substrate.

Hsie carried on further analysis of the development of resistance to antibiotics, using a bacterial strain (Mycobacterium ranae) that is related to the bacterium responsible for tuberculosis. He obtained additional evidence in support of the views that resistance is not induced by an interaction of the antibiotic with the bacterial cells, and that the pattern of development of resistance in a bacterial population is a property of the individual antibiotic to which it is exposed.

Several interesting observations were made by Wallace, King, and their group, working with irradiated populations. The first of these was that many lethals are eliminated from a population exposed to a single large dose of X-radiation. Those that are not eliminated, however, persist for a long time—more than 30 generations in these fruit-fly populations. That mutant genes are not to be regarded simply as deleterious genetic material was indicated by tests that estimated the well-being of the populations. These tests showed that individuals of the irradiated populations were at least as viable as individuals of the control population. Another interesting result was the determination of the amount of gamma-radiation (radium rays) required to form lethal genes in a population at twice the normal rate; 300 roentgens per generation were required. It is interesting (and speculative, at the moment) to compare this amount of radiation with the

maximum tolerance dose allowed by the Atomic Energy Commission—0.3 roentgens per week, or 390 roentgens in a 25-year generation. This comparison has little meaning at the present time, because only small fractions of the human population are exposed to any radiation, and these for relatively short periods of time. Of local interest was an analysis of populations of fruit flies living in stores or garbage pits in nearby localities. The frequency of lethal gene mutations in these natural populations was over 25%—higher than in many of the experimental populations. Further analyses of these populations are contemplated, with a view to obtaining data on chromosomes from populations much older than those being studied in the laboratory.

Again this year the largest group among the summer investigators was interested in research with microorganisms—namely, bacteria, bacterial viruses, fungi, and protozoa. Mark Adams, of New York University, experimented with crosses between two distinct bacterial viruses, and established a new serological group of phages. R. D. Hotchkiss, of the Rockefeller Institute, studied the transformation of rough-type pneumococcus cells into smooth-type under the influence of the purified specific nucleic acid from smooth cells. Together with his colleague, S. Granick, he tried unsuccessfully to induce the formation of a specific enzyme in a bacterium. Eva R. Sansome, of University College, Ibadan, Nigeria, conducted experiments designed to test whether giant forms could be produced in the bread mold Neurospora by treatment with camphor. The tests were not completed during the summer, but the preliminary experiments indicated that it may be possible to obtain these forms in Neurospora, as in Penicillium. I. A. Tittler, of Brooklyn College, studied the effect of aureomycin on the growth of protozoa (Tetrahymena), in the hope of discovering the mechanism responsible for increased growth in chicks when aureomycin is added to their diet. He found that growth in Tetrahymena was increased also. E. Racker, of New York University College of Medicine, purified from a bacterial extract an enzyme that catalyzes desoxypentose phosphate, thus making another step forward in his attempt to study the intracellular synthesis of desoxypentose nucleic acid. A. W. Bernheimer, of New York University, and E. Caspari, of Wesleyan University, obtained evidence that under certain conditions the caterpillars of the Royal Walnut Moth develop substances in their blood that exhibit antibody-like activities. In addition to this work, Caspari studied the development of a short-tailed strain of mice. H. A. Abramson continued to investigate lung function in normal and asthmatic subjects, and found that para-amino-hippuric acid is a suitable substance for the precise study of lung clearance of aerosols. Ernst Mayr, of the American Museum of Natural History, and D. Shemin, of Columbia University, spent the time in writing. During a large part of the summer the Laboratory enjoyed the stimulating presence of Sir Frank M. Burnet, director of The Walter and Eliza Hall Institute of Medical Research, Melbourne, Australia, and Professor F. W. Sansome, of University College, Ibadan, Nigeria.

Symposium

"The Origin and Evolution of Man" was the topic of the 1950 Cold Spring Harbor Symposium on Quantitative Biology, which was held from June 9 to June 17. About 120 scientists from various parts of the country and abroad attended the meetings, held in Blackford Hall from 9:00 to 4:00 each day. Thirty-seven speakers gave papers, and their results and interpretations were discussed by the entire group. This was the fifteenth session of the Symposia, which are sponsored by the Laboratory in order to bring together research workers in different but related sciences, and to stimulate cooperation among them.

The two main groups brought together by this program were geneticists and anthropologists, both of whom study the evolution of man. The ultimate purpose of science, perhaps, is to assist man in understanding himself and his place in the universe. All branches of science, even those concerned with celestial bodies and those studying the structure of the atom, eventually contribute to this purpose. But the sciences most immediately concerned with man are anthropology and biology, which study him as a living organism and a social being. For nearly two centuries anthropology and biology have developed almost independently, although both sciences have been profoundly influenced by such fundamental discoveries as Darwin's theory of evolution and his finding that man is a part of nature. In our century, the development of genetics, which studies the phenomena of heredity and variation, has caused a gradual drawing together of biological and anthropological research.

Man, like any other living organism, is a product of his heredity and his environment. Neither the heredity nor the environment can be ignored if we wish to arrive at a coherent understanding of an individual human being, or of a group of humans such as a population or race. Nevertheless, until recently, there has been relatively little contact and collaboration between anthropologists and geneticists or other biologists. The chief aim of the fifteenth Symposium was to help establish such collaboration.

The group started its nine-day session by considering the nature of the units of study with which anthropologists and biologists are concerned. The primary units that we observe are, of course, living individuals. These individuals, however, form natural units of higher orders—populations or groups, united by common descent, within which, rather than between which, marriages take place. Populations are of different orders. One of the largest of these is the human species, or mankind. Mankind is divided into races, subraces, and social, economic, linguistic, and other more or less clearly defined groups, each possessing its own heredity, or "gene pool." The human species has descended from pre-human ancestors, which, in some respects resembled extant apes and monkeys. Much progress has been made in recent years in studying these pre-human and early human forms, owing to the discovery of a large number of very in-

teresting fossil remains in several countries, particularly South Africa. This evolutionary development of man was discussed in two daily sessions of the Symposium.

A topic that interests both anthropologists and biologists equally is the genetic nature of traits that distinguish individual humans, or human populations such as races. Three sessions of the Symposium were devoted to analysis of human traits. Both "normal" traits, such as blood groups and characteristics of teeth, hair, and skin, and "pathological" traits, such as various hereditary diseases, were considered. This was followed by a discussion of the concept of race. Many misconceptions exist in people's minds about the nature and significance of racial variations in the human species. Considered scientifically, the problem is simple enough; different human populations differ in the frequency of certain genes in their hereditary constitution. The same genes that distinguish human races may also distinguish individuals within a race. Race differences are not absolute but relative.

The Symposium concluded with a discussion of the more general and philosophical implications of the modern Science of Man, and of the unsettled problems awaiting further study, in which anthropologists and geneticists will cooperate.

Participants in the meetings came from many parts of the United States, and from Puerto Rico, Denmark, England, Germany, India, Italy, Japan, and Sweden. The five foreign speakers on the program, invited to this country especially for the Symposium, were from Denmark, England, Italy, and Sweden.

Phage Meeting

It is a well-established custom for workers studying bacterial viruses (phages) to keep in close touch with one another, and to hold frequent conferences for discussion and evaluation of current research. Such a conference was organized last summer by Dr. Max Delbruck and held at the Laboratory on August 21 and 22. It was attended by about 35 people, approximately half of whom came from a distance.

Teaching

The Nature Study Course was given by Dr. and Mrs. J. Southgate Y. Hoyt of the Laboratory of Ornithology, Cornell University. They were assisted by Martin Pierce and Susie Mayr. This course is designed to stimulate interest in nature among the young people of the community, by showing them how to observe the many plants and animals around them, by teaching them how to answer the questions raised by their observations, and by helping them realize that careful and accurate study of the smaller incidents we all observe contributes greatly toward expanding our knowl-

edge of natural phenomena. The course was divided into three sections, according to the ages of the pupils, and was attended by forty-nine 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 sixth consecutive year an intensive three-week 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 College of Medicine, and had a capacity enrollment of sixteen students. A series of ten special seminars was arranged in connection with the course.

A three-week course in bacterial genetics, taught by Drs. E. M. Witkin, V. Bryson, and M. Demerec, was inaugurated this summer.. The purpose of the course was to give students a working knowledge of some of the techniques used in research in bacterial genetics, and to acquaint them with recent results of such work. The enrollment was twelve; and eight special lectures and demonstrations supplemented the regular program of the course.

Lectures

Weekly 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 the arrangements were made by Dr. Evelyn M. Witkin of the Carnegie staff. The speakers and titles were as follows:

July 20: Ernst Caspari, Wesleyan University. The origins of American scientists.

July 24: J. Southgate Y. Hoyt, Cornell University. Machias Seal Island—an island adventure.

August 3: N. Visconti di Modrone, University of Milan. Kinetics of the lethal action of nitrogen mustard on bacteria and phage.

August 10: S. Granick, Rockefeller Institute for Medical Research. Biosynthetic chains, chlorophyll, and evolution.

August 17: Eva R. Sansome, University College, Ibadan, Nigeria. "Gigas" forms of Penicillium obtained after camphor treatment.

August 24: Ernst Mayr, American Museum of Natural History. Polymorphism, especially in birds.

Special Events

Two illustrated lectures of general interest were given for members of the Association and their friends. On July 27, Sir Frank M. Burnet, the director of the Walter and Eliza Hall Institute of Medical Research in Melbourne, Australia, spoke about "Australian Work on German Measles (Rubella) in Pregnancy." He discussed research carried on by him, his collaborators, and other Australian scientists to discover the effects produced by the Rubella virus on human embryos. Another general lecture was given by Professor F. W. Sansome, head of the Botany Department at University College, Ibadan, Nigeria, on "Education and Life in Nigeria." Professor Sansome, formerly of the University of Manchester, England, joined the staff of the University College when it was organized in 1947, and has taken an active part in its development. He told a fascinating story of his life in Nigeria and his experiences with native students.

A demonstration of the research being done at the Laboratory and at the Department of Genetics of the Carnegie Institution was held in Blackford Hall on Sunday afternoon, September 24, and was well attended by members and friends of the Association. Scientists from the two laboratories explained the exhibits and discussed their work with the guests. The serving of tea and refreshments was efficiently organized by Mrs. Charles P. Noyes, Entertainment Committee Chairman of the Women's Committee.

Dining Room

The Blackford Hall dining room was in operation from June 7 to September 6, 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 about ninety persons, and during the remainder of the summer to between sixty and seventy persons. Mrs. Lillian Yongen acted as dining-room manager.

Laboratories and Equipment

The most significant changes made in the laboratories during the year consisted in additions to the equipment of the microbiology laboratory, primarily for use in teaching the courses on bacterial viruses and bacterial genetics. The equipment purchased included an incubator, several constant-temperature water baths, counting instruments, and a considerable quantity of glassware. In the Jones Laboratory a hot-water system was installed and an autoclave brought in, so that this laboratory may now be used for microorganism research, in which sterilization of materials is an essential requirement.

Buildings and Grounds

During the year an extensive program of modernization of residences, kitchen, and dining room was begun, utilizing the fund made available for that purpose by the Rockefeller Foundation. Mr. Francis Lowell, our new superintendent of buildings and grounds, is in charge of this work. To those familiar with the laboratory grounds, the most apparent change will be the removal of the cabins that stood in the field east of Blackford Hall to a clearing on the wooded hill west of Williams House. The ten cabins have been arranged in units of two or three and remodeled as four summer cottages, each including a living room, one or two bedrooms, and a bathroom with hot and cold running water.

The second major improvement that will be completed for use in the summer of 1951 is the modernization of the kitchen and dining room. A dishwashing machine, walk-in refrigerator, potato peeler, and grill have been installed in the kitchen. For the dining room we have obtained new tables and chairs, plastic dishes, and stainless steel utensils.

The third major improvement is the remodeling of the basement floor of Hooper House. This floor now has four large bedrooms and three baths, and will be equipped for heating during the winter months.

Finances

The expenses of the laboratory fall into two well-defined categories: running expenses (administration, operation, and summer activities), and expenses of the full-time research program. The first of these is covered by the dues and contributions of members of the Long Island Biological Association, the contributions of the Wawepex Society, the interest on securities, and the income connected with various Laboratory activities (rentals, overhead of research projects, course fees). This group of expenses includes the upkeep of buildings and grounds, an item for which the need is especially great; and in order to meet it our present income will have to be increased.

The expenses of full-time research are met by grants received from the Jane Coffin Childs Memorial Fund for Medical Research, the National Tuberculosis Association, the Army Chemical Corps, and the Atomic Energy Commission.

In 1950 the Symposium expenses were partially covered by the grant from the Carnegie Corporation for that purpose; and the Laboratory received a special grant from The Viking Fund for the expenses of participants from Scandinavian countries. A portion of the Polio Basic Research Fund of Long Island, which was raised through the initiative of Mrs. Lewis E. Pierson, Jr., was used for equipment needed for research with bacterial viruses.

Of great help to the Laboratory was the grant received from the Rockefeller Foundation for improvements to buildings and grounds.

Acknowledgments

It gives me great pleasure to acknowledge the support given 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; 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 Entertainment Committee, with Mrs. Charles P. Noyes as chairman, provided the refreshments for the Open House Tea and Exhibit in September.

Acknowledgment is 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 painting the John D. Jones Laboratory Building.

The Laboratory recognizes with gratitude the research grants made by the National Tuberculosis Association, the Army Chemical Corps, and the Atomic Energy Commission. The grant of the Carnegie Corporation again provided funds for expenses of the Symposium. These were supplemented by a grant received from The Viking Fund, Inc.

With much pleasure I acknowledge the interest shown by the Carnegie Corporation of New York and the Rockefeller Foundation in the work of the Laboratory, as expressed by their recent grants for construction of a lecture hall and for improvements to buildings and grounds.

M. Demerec
Director of the Laboratory

REPORTS OF LABORATORY STAFF

Morphological and Biochemical Analysis of Resistant Bacteria

V. Bryson, B. Prytz, H. Cuneo, E. Yongen, and C. Raymond

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Three fundamental questions continue to form the basis of our investigations of bacterial variability: How may strains of bacteria resistant to chemical and physical agents best be isolated? What are the biochemical properties of these resistant strains? What differences can be detected in the variability of resistant strains, in terms of modified mutation rate or altered genetic stability in the presence of mutagenic agents? Using isolation in agar or liquid media, numerous consecutive transfers have been completed of Escherichia coli grown in the maximum tolerable concentrations of mercuric chloride, dinitrophenol, sodium arsenite, cobalt chloride, proflavine, iodoacetamide, streptomycin, urethane, copper chloride, potassium tellurite, iodoacetic acid, phenyl mercuric nitrate, caffeine, sodium selenite, chloromycetin, cadmium chloride, uranylacetate, potassium cyanide, and malachite green. Most of the chemicals chosen are among those whose mode of action has already been explored by biochemists.

One result of these experiments has been the frequent induction of modified colonial morphology of E. coli plated on MacConkey agar, seen either as an alteration of the entire colony, or as an induced instability resulting in the appearance of numerous papillae and sectors within the colony itself, as described by Braun and Lewis for B/r. Since the parent strain B is relatively stable on MacConkey agar, the emergence, after chemical treatment, of an unstable type resembling B/r could be due to selection of spontaneously occurring B/r mutants normally present in large populations of strain B. This implies a greater resistance of B/r to the action of numerous toxic chemicals, and increased resistance of cells to radiation as a result of exposure to these same chemicals. Patterns of cross resistance are now under investigation and will show where a more critical test of the B/r selection hypothesis is required.

One may inquire to what extent the unstable colony type described by Braun and Lewis as characteristic of radiation-resistant B/r is an inseparable property of the strain. As an incidental procedure in an experiment, 35 low-grade lactose fermenters derived by ultraviolet irradiation of B/r (approximate dose 2000 ergs per square millimeter) were streaked on MacConkey agar and compared after two days of incubation at 37° C. As controls, streaks were made of strains 17S, B/r, B, and K-12. Observation of the bacterial populations showed many with unique morphology or with a relatively pronounced inhibition of one of the four or five morphologically distinct variants characteristically appearing on the "sandy" parent type of B/r. The most interesting variations were differences in

the stability of individual strains, ranging from the B/r colony type, which as previously stated is covered with papillae and sectors, to the stable B type. Two questions were raised. Is a reversion from B/r to B colony type associated with a reversion to radiation sensitivity; and, are the radiation-induced differences in degree of dissociation on MacConkey inherited?

Five of the radiation-induced mutants, together with B, K-12, and B/r strains, were grown in nutrient broth from a small inoculum and tested for sensitivity to radiation from a mercury vapor lamp. Simultaneously the broth-grown cells were tested for morphology on MacConkey. The greater radiation resistance of the B/r strain, from which five representative experimental mutants were chosen, was not modified by the reversion to B colony type. The modified colony types were inherited, at least through the fifteen generations in broth preceding plating. Morphological subtypes could be isolated by selection and growth of specific variants from the polymorphic colony in nutrient broth. Assay of such broth-grown cultures showed differences in the stability of subtypes. For example, strain K-12 plated on MacConkey showed occasional light sectors among the relatively stable dark colonies. Inoculation of dark colonies into broth, and growth, produced a population yielding only dark colonies on a dilution plating of approximately 200 colonies. Dilution plating on MacConkey of the culture inoculated with cells from the lighter areas gave 18% dark colonies, the remainder resembling the inoculum.

A complete analysis was made of strains obtained by exposure of E. coli to a minimum of ten transfers in nutrient-agar medium containing one of the following chemicals—mercuric chloride, sodium selenite, cobalt chloride, copper chloride, proflavine, sodium dinitrophenol, and crystal violet—and also of strains transferred for ten subcultures in nutrient broth containing either mercuric chloride, iodoacetic acid, sodium selenite, cadmium chloride, potassium tellurite, iodoacetamide, or proflavine. Each strain was streaked on MacConkey agar, and the predominant variant types were selected and retested for continued variation or stability. Further subdivisions were made until eighty-three strains had been established on the basis of origin. Each of these was inoculated into broth, cultured on MacConkey when grown, transferred through five subcultures in broth from the original liquid culture, and retested. As an example, we may consider the strain isolated after ten subcultures in nutrient agar containing partially inhibitory concentrations of crystal violet. Two main types of cell populations were observed on the MacConkey streaks prepared from this strain: a non-elevated sandy type, tending to produce numerous sectors and papillae, and a darker, elevated, more stable, semi-sandy type. From these, five additional types were obtained as further variants, and the variability of each of the five types was analyzed. The first four came from the non-elevated cell masses of the original plate: (1) a small, pink sandy variant, yielding on further test a type like itself and a larger colony type of a milky-brown color; (2) a large, pink sandy variant with irregular border, yielding on test six types—namely, small pink, others like itself (type 2),

a large milky-brown type, a dark colony type with transparent border, a lactose-negative variant, and a pale brown colony type; (3) a large brown variant with irregular border, yielding its own type and smaller round colonies; (4) a small brown variant producing its own type and larger irregular colonies (hence no genetic distinction between 3 and 4). From the original elevated semi-sandy cells the same elevated type was obtained, together with a few much darker colonies with irregular borders. The same general kind of analysis was performed with all the strains chosen for study.

It can be demonstrated from these experiments that the degree of genetic instability is a property of strains, as shown by a comparison of highly variable with stable types. Furthermore, the degree of stability can be modified by toxic chemicals and mutagenic agents, either toward greater variability (proflavine-treated B), or toward greater stability (ultraviolet-treated B/r). That differences in stability are the property of single cells within the same culture is suggested by a comparison of single colonies derived by dilution platings of the iodoacetic acid-treated E. coli. Stable colonies are observed adjacent to other colonies of the most highly variegated type. This means that some cells within a strain may reproduce in a stable manner, whereas others give rise to various biotypes, differing in mutability or selective value. The tendency of some types of cells to give rise to two or more subtypes makes qualitative experiments possible in liquid media.

Although the tendency of a colony to develop local areas that are morphologically and genetically distinct has been interpreted as a manifestation of genetic instability, these experiments do not distinguish between increased mutation and modified selective pressure as the cause of colonial variegation. In other words, the ultimate appearance of a papilla or sector within a colony depends not only on the mutation frequency, but also on the relative selective value of mutant and parent cells. Both phenomena may be subject to genetic modification and are ultimately allied with the changing cultural conditions of the environment. The possibility that the greater morphological variability of E. coli strain B/r (in contrast with strain B) is due to some form of segregation is of particular interest when a relatively multinucleate condition is hypothesized as the cause of radiation resistance. The genetic buffering effect of diploidy or polyploidy is well known. Aside from cytological evidence, which is difficult to interpret, the principal arguments against a polyploid basis for radiation resistance in B/r are the radiation resistance of most other E. coli strains (including K-12, a known haploid), the failure of B/r exposed to ultraviolet radiation to be sterilized at rates defining a multiple-hit curve, and the increasing evidence that radiation damage is primarily physiological rather than genetic. The additional finding that it is possible to isolate irradiated B/r which are relatively stable on MacConkey also suggests that the polymorphism of B/r is not necessarily an inseparable consequence of mutation to radiation resistance.

Previous knowledge of the effects of ultraviolet radiation on survival of E. coli subsequently grown in media containing dihydrostreptomycin has been increased by further studies. Since this subject formed the main thesis of our last report, we will summarize the present status of investigations on streptomycin resistance. Briefly, the following relationships appear to prevail when E. coli is grown in nutrient agar containing ten micrograms of streptomycin per milliliter. (1) The number of colonies obtained in streptomycin agar is positively correlated with the number of cells plated, up to a concentration of approximately 10^8 cells per ml; above this the number of colonies is reduced relatively and absolutely. (2) Increase in the survival level in 10 micrograms/ml streptomycin medium may be obtained by simultaneous addition of many carbohydrates at 0.3% concentrations, or at lower concentrations (0.03%) if the cells are briefly irradiated with ultraviolet in doses ranging from the 50% to the 1% survival level for strain B/r. (3) Most of the surviving colonies in both irradiated and nonirradiated populations are composed of mutants; however, the additional mutants seen after irradiation are not induced by ultraviolet, but rather are enabled as a result of treatment to survive at the competitive pressure of high population density. (4) Ultraviolet irradiation produces its final effect directly on the cell, and does not merely antagonize streptomycin through an indirect inactivation of the antibiotic when irradiated cells are added to streptomycin medium. (5) The enhanced survival of E. coli in streptomycin-carbohydrate medium after ultraviolet treatment is most marked if the essential trace of carbohydrate is of a type normally metabolized by the organism, and if the added carbohydrate has been employed as sole carbon source during growth of the cells before experimental use. An attempt is now being made to interpret these observations on the basis of the structural similarity of portions of the streptomycin molecule to hexoses.

A study has been carried out in the past year on the action of streptomycin on the E. coli K-12 strain, and on some of its streptomycin-dependent mutants. A report by Umbreit and his co-workers indicated that the action of streptomycin on E. coli is associated with the citric acid cycle, through a direct inhibiting action on oxalacetic acid condensation, which prevents the first step in the tricarboxylic acid cycle.

Since streptomycin-dependent mutants of E. coli, as well as the parent streptomycin-sensitive organism, were available, a detailed study of this reaction was undertaken. A number of respiratory studies was carried out with the Warburg respirometer, using the streptomycin-sensitive K-12 strain and two dependent strains. The cells were grown in a complete broth medium, harvested by centrifugation, washed twice with saline solution, and finally resuspended in saline solution in such a concentration as to contain 0.5 mg bacterial nitrogen per ml of suspension.

Oxalacetic acid in solution is an extremely unstable substance; it decarboxylates spontaneously to form pyruvic acid and carbon dioxide. This reaction can be accelerated by a decarboxylase present in E. coli. Pyruvic

acid is probably the compound that takes part in the condensation reaction, although it is possible to visualize a momentarily reactive product originating from oxalacetic acid to form the first step in the tricarboxylic acid cycle. However, since the reaction leading from oxalacetic acid to pyruvic acid is reversible, both compounds may have a part in it. Pyruvic acid is in turn oxidized to form acetic and formic acid, or acetic acid and carbon dioxide. This acetic acid may be activated through a phosphorylation mechanism so that it can condense with other active compounds present, or it may undergo an oxidation-reduction reaction and be dehydrogenated to produce succinic acid. The formic acid derived from the oxidation of pyruvic acid is oxidized to carbon dioxide and hydrogen, which in nascent form may react with any hydrogen acceptor present.

In an experimental setup in the Warburg respirometer, formate and pyruvate, singly and combined, both with and without streptomycin, were added to bacterial suspensions of different strains. It was found that very little oxygen consumption took place in the vessel containing the streptomycin-sensitive K-12 strain on a formate-and-pyruvate substrate, whereas the dependent strain showed very high oxygen consumption under the same circumstances. Furthermore, it was shown that the presence of streptomycin did not affect the oxidation of either formate or pyruvate in any of the strains.

When the same experiment was carried out under anaerobic conditions, and the carbon dioxide liberated was measured, no such difference was found except that streptomycin appeared to stimulate the hydrogen transfer from formate to pyruvate, to produce lactic acid. This was in accord with the experiments on aerobic oxidation. As hydrogen produced from the oxidation of formate is removed by the hydrogen acceptor, pyruvate, the equilibrium shifts and more hydrogen is formed.

The same type of result was obtained when oxalacetic acid was used instead of pyruvate. There is a stimulation of oxalacetate oxidation in the presence of formate, and a still greater stimulation in the presence of formate and streptomycin. The same type of reaction may take place here; that is, streptomycin may accelerate hydrogen transfer to the keto-acid acceptor.

These results indicate that the oxidation-reduction mechanism, rather than oxalacetic acid condensation, is involved in streptomycin resistance or dependence.

Further evidence that the condensation reaction and the subsequent tricarboxylic acid cycle does not take place at all, or is not important, in E. coli may be seen in the fact that no tricarboxylic acid or derivative could be found on a paper cromatogram developed from a metabolite mixture consisting of oxalacetic acid and pyruvic acid. Furthermore, citric acid was oxidized only very little or not at all in a Warburg respirometer by either a streptomycin-sensitive or a streptomycin-dependent strain. Finally, no evi-

dence of a citric acid dehydrogenase could be found in any of the strains, using either triphenyltetrazolium or methylene blue as the indicator.

It appears that hydrogen transfer from a donator to an acceptor, possibly a keto acid, is involved in streptomycin resistance. A direct proof that such a reaction does take place was obtained by a determination of lactic acid developed in a substrate containing either oxalacetic acid or pyruvic acid. Utilizing pyruvic acid in the presence of streptomycin, 72 micrograms of lactic acid per ml of reaction mixture were formed in 30 minutes, and in 60 minutes this had decreased to 48 micrograms. In the absence of streptomycin, the figures were 55 and 48 micrograms per ml at 30 and 60 minutes. In the presence of streptomycin more lactic acid was formed—that is, hydrogen transfer was activated and also lactic acid metabolism was stimulated. If a hydrogen acceptor more active than pyruvic acid was present in the system, less lactic acid was formed. When triphenyltetrazolium hydrochloride was added to the system, for instance, only about 1 microgram of lactic acid per ml of reaction mixture could be found.

The reactions described above appeared to involve irreversible hydrogen transfers, and it is not very likely that a reversible hydrogen-donator-acceptor, like coenzyme I or II, was involved. As hydrogen donators one may consider formic acid and acetic acid, both originating from pyruvic acid. It was demonstrated that acetic acid can be dehydrogenated to form succinic acid, identified by paper chromatography. The tricarboxylic acid cycle can thus be detoured through this reaction and the metabolic path can go directly from acetate to succinate and through the rest of the cycle back to oxalacetate and pyruvate.

In all the dehydrogenase experiments it was evident that a much larger "blank" value was found for the streptomycin-dependent strains than for the streptomycin-sensitive strain. The average "blank" values found for the sensitive strain and a resistant strain were 27 and 245 micrograms of formazan produced per mg of bacterial nitrogen per hour, respectively. This fact may indicate that the dependent strains possess a higher content of either coenzyme I or coenzyme II, or both. A spectrophotometric determination of reduced coenzyme measured at 340 millimicrons showed a consistent but small increase of coenzyme content in the dependent strain as compared with the sensitive parent strain. However, this method does not differentiate between coenzymes I and II. An enzymic method specific for coenzyme I was then used. Here diaphorase, lactic apo dehydrogenase, and lactate were employed, and the limiting factor was the coenzyme content of the standardized cell suspension. By this method it was shown that the dependent strains contained a considerably larger amount of coenzyme I than the sensitive strain.

A relation was thus demonstrated between streptomycin resistance or dependence, coenzyme content, and—probably—hydrogen transfer. An attempt was made to see whether the sensitive strain could be grown in the

presence of streptomycin upon the addition of coenzyme I or fractions thereof. Irrespective of the dose, up to 100 micrograms of coenzyme I per ml of complete medium, no growth of the sensitive strain could be obtained in the presence of streptomycin. The same was true when either nicotinic acid or nicotinic acid amide was used. Neither adenine nor ribose had any effect. However, in several experiments, growth of the sensitive strain in the presence of streptomycin was obtained upon the addition of fumarate. This appears to be a specific effect, for hydrogen acceptors like triphenyltetrazolium hydrochloride or methylene blue did not act similarly. Fumarate, of course, is a natural metabolite whereas the two dyes used are both foreign materials. It is not possible to attribute the antagonistic effect of fumarate assuredly to its role as a hydrogen acceptor, since many substances have been shown by other workers to antagonize the effects of streptomycin through a mechanism that does not appear to involve hydrogen transport.

Effects of Chemotherapeutic Agents and Surface-Active Chemicals on Mycobacteria

Jen-yah Hsie

By employing a specially devised cylinder-plate method to differentiate various grades of dihydrostreptomycin-resistant and -dependent bacterial strains, it has been possible to approach the problem of the origin of drug resistance and dependence by means of the following reasoning. If the emergence of drug-resistant and -dependent strains is due to interaction of the drug and the sensitive cells, then different concentrations of the drug should have different influences on the sensitive cells, and result in the emergence of different numbers of resistant and dependent cells. At first glance, the data obtained might appear to indicate that such is really the case, because the total number of resistant cells obtained when a large sensitive population is treated with dihydrostreptomycin is a function of the drug concentration. However, a further analysis of the degree of resistance and dependence, by the simple differentiation method, reveals that the number of mutants resistant to more than 1000 units of dihydrostreptomycin, or dependent on streptomycin for growth, is about the same regardless of the drug concentrations employed in their isolation. Therefore it appears that these fully resistant and dependent individuals are not products of an interaction of the drug with the sensitive cells, since higher concentrations of the drug do not result in higher numbers of mutants.

As pointed out in the previous Annual Report, the development of resistance to neomycin and choloromycetin in Mycobacterium ranae follows an obligatory multiple-step pattern, and resistance to streptomycin a facultative multiple-step pattern. With a nonpathogenic human strain of the tubercle bacillus (H37Ra) the same patterns were observed, providing additional evidence that the resistance pattern most probably is a property of the individual antibiotic. Thus, when exposed to a specific antibiotic, unrelated strains of bacteria differ in degree of resistance, but tend to possess the same qualitative type of resistance pattern.

Resistance to zephiran chloride, sodium para-amino salicylate (NaPAS), and terramycin was built up experimentally in five, three, and two steps, respectively, by selection and subculture of survivors in gradually increasing concentrations of the drugs. But when these resistant strains were subcultured in plain broth, they gradually shifted back in the direction of sensitivity, unlike the strains resistant to other drugs discussed above. The step-wise increment of resistance to these three chemicals in the presence of gradually increasing concentrations appeared to be linear, instead of exponential as in the development of resistance to streptomycin, neomycin, and chloromycetin.

Experiments on growth rates and competitive growth of the various resistant and dependent strains, in the presence and in the absence of the drugs, contributed to an understanding of population shifts from sensitivity to resistance and dependence.

It was found that the mean generation time of the wild-type strain of M. ranae in plain Triton A20 nutrient broth was about 2.1 hours; and that of a highly streptomycin-resistant strain, either with or without streptomycin, was 2.9 hours. The mean generation time of a strain slightly dependent on streptomycin was 3.3 hours in 1000 micrograms of streptomycin per milliliter of broth, 3.8 hours in 10 micrograms per milliliter, and 4.1 hours in broth containing only traces of streptomycin. Comparison of growth rates in mixed cultures was possible because individual strains may be identified by type of colony formed on glycerolated nutrient agar containing either no streptomycin or 1000 micrograms per milliliter. The order of growth rates among the various sensitive, resistant, and dependent strains, beginning with the highest, is as follows: streptomycin-sensitive, highly streptomycin-resistant, slightly streptomycin-dependent, highly streptomycin-dependent.

Under identical conditions, repeated growth experiments with seven cultures of the wild-type strain of M. ranae and five cultures of the third-step NaPAS-resistant and fifth-step zephiran chloride-resistant strains showed that the mean generation time of the first is 2.1 hours, with a standard deviation of ± 0.08; that of the other two, 2.5 ± 0.23 and 2.6 ± 0.11 hours, respectively. In the presence of sublethal concentrations of the drug, the lag period and mean generation time in the wild-type strain were greatly prolonged, whereas the resistant strains were less affected by similar concentrations.

Chemicals that only affect the phenotypic expression of the microorganisms and are not involved in qualitative or quantitative changes in the enzymatic system, were also tested. It was found that surface-active agents, such as Triton A20 and Tween 80, could convert all the R- and M-form colonies of M. ranae into S forms. On the other hand, glycerol and asparagine media could convert either S or M forms into R forms. These changes were transitory, noninheritable, and analogous to acquired environmentally conditioned differences in higher organisms.

To facilitate recognition of the more productive strains of SM-producing organisms, an identification method was devised. The different grades of SM productivity of the various normal and mutant strains of Streptomyces griseus can easily be compared by measuring the width of the growth zone of dependent mycobacterio grown in their presence. This method is convenient and reliable, and might aid in choosing the most productive strain for commercial use. It also makes the development of highly productive mutants by treatment with physical and chemical mutagens less laborious, by facilitating recognition of differences in antibiotic productivity among the mutants.

Radiations and Populations

Bruce Wallace and James C. King

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During the past year we continued our analyses of irradiated populations in an effort to determine to what extent, if any, the induced mutations modify the adaptive value (= well-being) of populations. The main effort dealt with chromosomal content of the populations—frequencies of lethals and semilethals among samples of chromosomes, and average effects of nonlethal, non-semilethal chromosomes on the viability of homozygous individuals. In addition, the first attempts were made to estimate experimentally the adaptive values of our laboratory populations. Finally, for purposes of comparison, 446 chromosomes from local wild populations of D. melanogaster were analyzed.

Three new experimental populations (Nos. 5, 6, and 7) were added to our studies. (Populations 1, 2, and 3 are described in the Annual Report for 1949.) Populations 5 and 6 received chronic gamma radiation from a radium source at a rate of 5.1 r per hour—nearly 2000 r every two weeks, the normal sample interval. These two populations differed in size; No. 5, which was kept on unyeasted culture medium, was limited in size to 100-1000 adult flies; No. 6, kept on medium enriched with brewers' yeast (as were the original populations), was maintained at approximately 10,000 adults. Population 7 (size, 10,000 adults) was exposed to a smaller radium source and received only 0.9 r per hour, or about 300 r during the sample interval.

From the data given in Table 1, it is evident that lethals accumulated much more rapidly in the irradiated populations than in the control (No. 3). In populations 5 and 6, lethals accumulated at a rate of 5% - 6% per two-week sample interval (no effect of the different population sizes was yet evident), and in population 7 at a rate of about 1% per two weeks. The rate at which lethals accumulated in the control population was 0.37% per sample interval.

Table I

Frequencies of lethals in samples of chromosomes taken from experimental populations of D. melanogaster

Sample	No. 1	No. 3	No. 5	No. 6	No. 7
1	18.3%	0.8%	9.1%	10.7%	2.7%
2	14.1	0	....	.....	....
3	11.6	2.2	14.2	19.0	4.2
4	10.1	....	.....	.....	....
5	.....	1.4	26.4	31.0	7.0
6	10.9	....	.....	.....	....
7	.....	4.6	23.6	31.3	5.8
8	13.7	....	.....	.....	....
9	.....	3.2	39.4	35.7	9.6
10	14.6	....	.....	.....	....
11	.....	4.9	.....	.....	....
12	14.5	....	.....	.....	....
13	.....	5.2	48.9	49.2	12.2
14	13.6	....			
15	.....	4.0			
16	18.8	....			
17	.....	6.1			
18	18.7	....			
19	.....	4.9			
20	13.8	....			
22	.....	7.3			
24	20.0	....			
26	.....	10.6			
28	25.4	....			
30	20.8	12.9			

A comparison of the rates of accumulation of lethals in populations 1 and 3 yielded information concerning semidominance of lethal genes—the degree to which lethals are manifested in heterozygotes. After the initial period of elimination of lethals from population 1, lethals increased in frequency at the rate of 0.47%±0.08% per sample; the comparable increase in population 3 was 0.37%±0.04%. The difference between these two rates is not significant. In order to determine the maximum amount of semidominance of lethals consistent with the data, it was necessary to determine the difference between the least possible increase in population 1 and the maximum possible increase in population 3; this difference is 0.17% per generation. In relation to the 8%-9% original difference between the frequencies of lethals in the two populations, this 0.17% represents a semidominance of 2%, the maximum possible semidominance of these lethals. As the number of samples increases, this maximum value will be ascertained with greater accuracy.

The average frequencies of wild-type flies in "normal" (nonlethal, non-semilethal) test cultures of different samples reflects the presence of subvitals (genes with extremely small deleterious effects) within the populations. Unfortunately, chance fluctuations of these values are large enough to necessitate a study of many samples in order to draw conclusions regarding such genes. At the present time, population 3 is the only one suitable for analysis (Table 2);

Average frequencies of wild-type flies in test cultures of nonlethal, non-semilethal chromosomes taken from experimentl populations of D. melanogaster

Sample	No.1	No. 3	No. 5	No. 6	No. 7
1	30.66%	32.08%	31.75%	31.43%	31.52%
2	31.94	31.65	.....	.....	.....
3	31.11	31.46	30.77	30.60	31.41
4	30.39	.....	.....	.....	.....
5	.....	31.49	31.39	30.55	30.81
6	30.79	.....	.....	.....	.....
7	.....	32.14	30.36	30.35	31.27
8	29.63	.....	.....	.....	.....
9	.....	31.64	30.31	30.99	31.42
10	29.85	.....	.....	.....	.....
11	.....	31.40	.....	.....	.....
12	30.81	.....	.....	.....	.....
13	.....	31.79	30.11	28.11	30.60
14	29.99	.....			
15	.....	30.84			
16	29.90	.....			
17	.....	30.87			
18	29.26	.....			
19	.....	32.25			
20	30.33	.....			
22	.....	32.77			
24	30.63	.....			
26	.....	32.04			
28	31.04	.....			
30	30.83	32.77			

the average viability of "normal" flies within this cage (indicated by the average frequency of wild flies in the test cultures) has remained constant—the regression (average change per sample) of the viabilities is .0001 ± .0001. This value, which is also a function of u, the mutation rate of subvitals, and s the average degree of subvitality, allows us to set reasonable values on u and s. In the case of the control population, for instance, the maximum downward slope of the regression of viabilities is—.0001; this indicates that former estimates of .016 for u and .15—.50 for s are too large and must be modified.

The data discussed above dealt with the genic or chromosomal content of the populations, and were essential for understanding changes that occurred within them. The adaptive value of a population, however, must be estimated by other means. An approach to the latter problem was made by utilizing matings that yielded cultures of genetically marked flies and wild flies carrying second chromosomes of separate origin. The combinations of wild cromosomes were chosen at random, and hence the series of test cultures in a given sample represented a cross section of the population at that time. A comparison of the results of tests of populations 3, 5, 6, and 7 led to estimates of adaptive values of 1.00 (the standard for comparison), .93, .96, and .98. It seems that the viability of flies from the irradiated populations was lowered—at least for populations 5 and 6 the ones that received the most radiation. However, if the result for population 3 is retained as the standard (1.00), the adaptive value of population 1 is 1.04; and the difference is probably significant. This indicates that the ebb and flow of genetic materials within successive generations of a population is an exceedingly complex process. In each population, chromosomes leading to well-balanced individuals are probably favored by natural selection over other chromosomes. The result in population 1 has been the selection of chromosomes that yield, on the average, individuals more viable than similar individuals from the control population, although, as is evident from Tables 1 and 2, individuals that are homozygous for chromosomes from this irradiated population are generally inferior to similar individuals of the control.

The amount of genic diversity present in natural populations is generally not appreciated. An indication of the possible diversity can be seen in the lethal data for the control population (Table 1); although the 30 generations included in this table were but a moment in the history of D. melanogaster, the population had already accumulated over 10% lethals. Throughout the ages that fruit flies have existed in natural populations, lethals have accumulated at rates comparable to the 0.4% determined by our study. After a sufficient time, however, lethals accumulate to such a point that their elimination counterbalances their formation. To aid in our understanding of the experimental populations, chromosomes from two local populations of D. melanogaster (A & P Food Store, Syosset, N. Y., and a compost pit, Huntington Station, N. Y.) were analyzed. The frequency of lethals in the different samples taken from these two populations did not differ significantly, and so the data could be consolidated; 113 lethals were recovered from 446 chromosomes studied; the frequency of lethals in these populations was 25.3%. The average viability of flies homozygous for normal chromosomes was 29.84%. The frequency of lethals found in natural populations of this fly is greater in more southern regions where the breeding season is longer and the size of the populations larger; Florida populations have lethal frequencies as high as 50-60%. Obviously, neither population 1 nor population 7 has accumulated mutant genes to the extent found in local Long Island populations; populations 5 and 6 have exceeded them,

however, and are more like those that might be found in Florida. A point that must be determined at some later time is the relative viabilities of flies carrying different chromosomes from natural populations; for, just as population 1 exceeded population 3, so might natural populations that have existed for long periods of time exceed the newly made experimental populations.

REPORTS OF SUMMER INVESTIGATORS

Abramson, Harold A., 133 East 58th Street, New York, N. Y. —— The study of lung function in normal and asthmatic subjects was continued by investigating the way in which inhaled mists of para-amino-hippuric acid diffused through the lungs into the blood stream. Since 90% of the material is excreted by the kidneys with each circulation of the blood volume, one can think of the kidney excretion as expressing the diffusion rate through the lungs. Confirming previous experiments with the negatively charged dye phenolsulfonphthalein, it was found that the excretion of para-amino-hippuric acid, in the range studied, was proportional to the inspiration time of the mist. Since several toxic reactions were experienced with phenolsulfonphthalein, and none with para-amino-hippuric acid, it appears that para-amino-hippuric acid is a suitable substance for the precise study of lung clearance of aerosols.

Adams, Mark H., New York University College of Medicine, New York, N. Y.——A bacterial virus attacking Salmonella paratyphoid B has been found to be serologically related to coli phage T5. It is indistinguishable from T5 morphologically and in sensitivity to ultraviolet light but differs significantly in host range, latent period, heat stability, and rate of neutralization by PB and T5 antisera. A common host for these two viruses was found in strain Cullen of E. coli. Both viruses multiply simultaneously in single cells of this bacterium, and under these conditions recombination of genetic characters occurs. The hybrid virus particles isolated from such mixedly infected bacteria differ from both parental types in heat sensitivity, resemble T5 in host range, and resemble PB in latent period. This extends a phenomenon originally discovered by Delbruck in the T2, T4, T6 serological group of coli phages to a different and unrelated serological group including PB and T5.

Bernheimer, Alan W., New York University College of Medicine, New York, N. Y., and **Caspari, Ernst,** Wesleyan University, Middletown, Conn.——In contrast to results obtained using mammals, experiments with caterpillars have shown that the tissues of caterpillars belonging to different genera and even to different families can be successfully transplanted. This remarkable tissue compatibility could be explained if caterpillars could be shown to lack the ability to form antibodies. We therefore undertook to examine for antibody-forming capacity the larva of the Royal Walnut Moth, Citheronia regalis, specimens of which weigh as much as 25 grams and yield as much as 1.5 cc of blood. The antigens employed were human erythrocytes of blood group O and

crystalline egg albumin. The bloods of several caterpillars that had previously been injected with erythrocytes were observed to produce hemagglutination in vitro in dilutions up to 1:20. Of the bloods of a considerable number of caterpillars not previously injected with erythrocytes, all but one failed to hemagglutinate. Results of a somewhat similar character were obtained with egg-albumin-immunized caterpillars. The findings suggest that, under certain conditions, the larvae develop in their blood substances that exhibit antibody-like activities. The data do not indicate that we are necessarily dealing with antibodies comparable to those elicited in mammals. And, in particular, there remains some doubt as to how the effects observed are specifically related to the nature of the materials injected. The results are of a preliminary nature, but we believe that they are of sufficient interest to warrant a continuation of the study.

Caspari, Ernst, Wesleyan University, Middletown, Conn.——Heterozygous T/+ mice have short tails. The development of this character, as described by Chesley (1935), consists in the formation of a thin distal part of the outgrowing tail in embryos 11 days old. This thin part at first contains somites, but at later stages it dedifferentiates and disappears. The developmental fate of the distal parts of +/+ and T/+ embryonic tails after transplantation into the anterior chamber of the eye of normal adult mice was studied. One hundred and twenty-six eyeballs into which pieces of tails from embryos 10-12 days old had been implanted 5 or 6 days before fixation were sectioned and stained with hematoxylin-eosin or with azan. The preparations were studied and the following results obtained. Distal pieces from +/+ and T/+ embryos developed equally well in the anterior chamber. Both types of graft developed cartilage, sometimes tubular structures, which may have been neural tube or tail gut, and abnormal structures of cystic character. The main difference between the two genotypes was that in +/+ distal tail pieces a clear notochord was always associated with the cartilage wherever cartilage is formed. In T/+ distal tail pieces cartilage was frequently formed, but in no case was any notochord found connected with it. Explants from the basal part of T/+ tails combined both cartilage and notochord, but the notochord appeared either interrupted or forked. Both these abnormalities have been described in the development of T/+ tails. Although the material is not sufficient to permit general conclusions concerning the manner of action of the gene T, certain facts seem to be indicated. (1) In explants, distal T/+ tail pieces survive and differentiate similarly to +/+ tail pieces. (2) No notochord appears in distal T/+ explants, whereas it is usually present in +/+ and proximal T/+ explants. It thus appears that the notochord may be the principal structure affected by the action of the gene T. The role of the notochord in the organization of the tail seems to be complicated, and will have to be investigated by further experiments.

Granick, S., The Rockefeller Institute for Medical Research, New York, N. Y.——Part of the work during the summer consisted of writing a review on the "Biosynthesis of Heme and Chlorophyll" for the Annual Reviews of Plant Physiology. Experimentally an attempt was made, in collaboration with Dr. Rollin D. Hotchkiss, to induce the formation of a specific enzyme in a bacterium by a transforming reaction with an appropriate desoxyribose nucleic acid. This failed, but it may have been due to the fact that the two organisms used were rather unrelated. The experiment was set up in the following way. A smooth hemophilus influenzae Turner, which requires heme for growth and cannot make its own heme, was treated under appropriate conditions with DNA isolated from H. parainfluenzae Latier, a rough organism. If a transformation had occurred, then H. influenza might have grown in the absence of heme and it might also have acquired the antigenicity of the rough H. parainfluenzae. Neither of these changes occurred.

Hotchkiss, Rollin D., The Rockefeller Institute for Medical Research, New York, N. Y.——Rough pneumococci growing in the presence of the purified specific nucleic acid from Type III encapsulated (S) strains are in part "transformed" inheritably into Type III S cells. Estimates of the proportion of S to R (rough) colonies present after six to eight hours of incubation had indicated one S colony for every six to twenty R colonies. Continuing this work at the Biological Laboratory, the ideal was more nearly approached of obtaining the S and R counts after a minimal exposure to transforming nucleic acid. Means were found, during the summer to estimate the S count produced during as little as five minutes' exposure of a 4-hour culture to the transforming factor. The S cells have a moderate selective advantage in the transforming environment on account of the anti-R agglutinins present in this system. Accordingly, the S count after this earlier, briefer exposure is much smaller, both in an absolute sense and in relation to the R count, than that mentioned above. Nevertheless, it is a striking fact that during five minutes' exposure to the nucleic acid 2000 to 3000 S cells can be produced per milliliter from a total population having only 100 to 200 times this many R colonies per milliliter. Longer times of exposure and higher concentrations of the nucleic acid transforming factor resulted in greater conversion to S, and some progress was made toward determining the concentration dependence of transformation. The quantitative characteristics of this dependence should reveal important clues to the theoretical mechanism by which inheritable changes are brought about in microorganisms through the action of nucleic acids. In experiments conducted with Dr. S. Granick, attempts were made to transform biochemical properties of the species Hemophilus influenzae.

Mayr, Ernst, The American Museum of Natural History, New York, N. Y.——Two projects were carried out during the summer. First, I completed the manuscript of a book entitled "Methods and Principles in

Taxonomy" (in joint authorship with Drs. E. G. Linsley and R. Usinger). Secondly, I made a study of the insect fauna found in mushrooms of the Cold Spring Harbor area. This study is now being continued by one of the students of Dr. Herman Spieth.

Racker, E., New York University College of Medicine, New York, N. Y.—— Bacterial as well as animal viruses contain desoxypentose nucleic acid (DNA) but little is known about the biosynthesis of desoxypentose sugar. Since a simple aldol condensation between glyceraldehyde phosphate and acetaldehyde would result in the formation of desoxypentose-5-phosphate, a search was made for an enzyme catalyzing this reaction. Such an enzyme was readily demonstrated in extracts of several bacterial species, and was purified from E. coli extracts. The purified enzyme catalyzes the reversible reaction between glyceraldehyde-3-phosphate + acetaldehyde and desoxypentose-5-phosphate. Desoxypentose phosphate is isolated as the end product of this reaction as a water-soluble, alcohol-insoluble barium salt. In the reverse reaction, desoxypentose phosphate, prepared from desoxy-adenylic acid by mild acid hydrolysis, is split by the same enzyme to triose phosphate and acetaldehyde. Since E. coli contains an enzyme which splits ribose-5-phosphate to triose phosphate, desoxyribose phosphate is formed when acetaldehyde and ribose phosphate are added to the two enzymes required for this transformation.——Two inhibitors capable of interfering with the enzymatic synthesis of desoxypentose phosphate in the extract are chloral hydrate and propionaldehyde.——It is hoped that with the help of these and similar inhibitors a study of intracellular synthesis of DNA can be made.

Sansome, Eva R., University College, Ibadan, Nigeria. —— In addition to work in the library, a piece of research was carried out on the effect of camphor treatment on Neurospora crassa. "Gigas" forms have been obtained after camphor treatment in various strains of Penicillium. However, since there is no known sexual stage in this organism it is not possible to determine their nature by direct genetical methods. An attempt is therefore being made to obtain such forms in Neurospora. The identification of "gigas" forms depends on the examination of spores of a uninucleate type. Consequently the fluffy microconidial strain of N. crassa, kindly supplied by Dr. Garnjobst, which has uninucleate microconida and no macroconidia, is being used in the experiments. Wide, slanted tubes containing malt agar and natural camphor, and control tubes without camphor., were inoculated with a suspension of microconidia before we left Africa. Growth was inhibited for some time in the camphor tubes, and, when it did occur, was stunted. When conida were formed in the camphor tubes, samples of conidia from the control and treated tubes were plated, and single-spore cultures isolated. When the single-spore cultures were sufficiently grown, samples of their conidia were

mounted in water, and camera lucida outlines were drawn at a magnification of ca. 1900. The drawings of the individual cultures were compared for conidial size. This examination of conidial size is tedious and difficult, because the conidia are much less abundant than in Penicillium, for instance, and there is a greater irregularity in conidial size. Nevertheless, determinations were made, as follows. Twenty-eight cultures from the control, of normal microconidial appearance, had conidia of approximately the same size as had one "fluffy" culture, which appeared presumably as a mutation at the microconidial locus. The 30 treated cultures were all of the microconidial fluffy type, and 23 had conidia of the same approximate size as the controls, whereas two (cultures T21 and T27) had larger conidia. T21 had a few conidia of standard size among the larger conidia. Two successive sub-cultures of T27 had larger conidia, whereas two subcultures of T21 had a mixture of larger and standard-size conidia. Crosses were made between control microconidia cultures and wild-type, between T21 and wild-type, and between T27 and wild-type. Preliminary observations indicated that the wild-type × T27 cross is very sterile whereas the wild-type × control cross produces some perithecia with asci. This is in accordance with the view that T27 is diploid. Attempts are now being made to obtain reasonably interfertile microconidal strains of both mating types, after which an effort will be made to obtain gigas forms in both mating types so that they may be intercrossed. I shall also try to obtain meiosis in the gigas standard cross. The preliminary experiments indicate that it may be possible to obtain gigas forms in Neurospora as in Penicillium.

Shemin, David, College of Physicians and Surgeons, Columbia University, New York, N. Y.——Since I had worked out, at Columbia University, the source of carbon atoms of porphyrins and the elucidation of some of the intermediates, I continued this line of research on chlorophylls at Cold Spring Harbor. I worked out methods of isolating chlorophyll and growing Chlorella. This was done in order to investigate the mechanism of chlorophyll formation. With the experience gained at the Laboratory, I was then able to continue the work at Columbia.——In the quiet atmosphere I was able to write a long manuscript for the Journal of Biological Chemistry, and to exchange viewpoints with many of the other people staying at the Laboratory. I presented an informal seminar on the status of one-carbon compounds.

Tittler, Irving A., Brooklyn College, Brooklyn, N. Y.——The recent work of Stokstad et al. (Proc. Soc. Exp. Biol. & Med., 1950, 73) indicated that aureomycin produced a growth response when fed to chicks on an otherwise adequate diet. Since these results may have been due to a direct effect of the aureomycin on the intestinal flora, it was decided to repeat the experiments on a pure culture of Tetrahymena, whose metabolism is strikingly similar to that of higher organisms. This would serve the dual purpose of determining whether the aureomycin effect is a truly

proliferative one on cells as such or actually operates in some other, indirect manner. Preliminary runs on Tetrahymena, using various concentrations of yeast extract and aureomycin, indicated that aureomycin concentrations higher than 1.0 mg % were toxic. Increases in cell population were noticed in cultures grown in 5% yeast extract with 1.0 mg % added aureomycin. Using chemically defined media without added aureomycin, growth was uniformly poor unless a small quantity (0.01%) of alkaline hydrolyzed yeast nucleic acid was added. However, additions of small amounts (less than 1.0 mg %) of aureomycin (99.3% pure) resulted in heavy growth in the absence of any added hydrolyzed yeast nucleic acid. The exact mechanism of this effect is still unknown and is being further investigated.

COURSE ON BACTERIOPHAGES

June 26-July 15, 1950

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

Assistant: Maryda Swanstrom, New York University.

An intensive three-week course dealing with techniques and current research developments in the field of bacterial viruses was given for the sixth year. A new experiment, involving the photoreactivation of ultraviolet-inactivated viruses, was added to the schedule. Sixteen students were enrolled in the course; eight were research workers with the doctoral degree, and most of the remainder graduate students working for the doctorate. The students are listed below:

Guy T. Barry, Ph.D., Organic Chemistry, Rockefeller Institute, New York City

William Belser, Graduate Student, Microbiology, Yale University, New Haven

H. T. Epstein, Ph.D., Physics, University of Pittsburgh

Lorraine Gosselin, Technician, Rockefeller Institute, New York City

N. Grossowicz, Ph.D., Microbiology, University of Jerusalem, Palestine

P. E. Hartman, Graduate Student, Microbiology, University of Pennsylvania, Philadelphia

Ruth F. Hill, Graduate Student, Biophysics, Columbia University, New York City

Jen-yah Hsie, Ph.D., Microbiology, Biological Laboratory, Cold Spring Harbor

Mary Lanning, Technician, Children's Hospital, Philadelphia

Robert Loevinger, Ph.D., Physics, Mt. Sinai Hospital, New York City

Lionel A. Manson, Ph.D., Biochemistry, Western Reserve University, Cleveland

Noel R. Rose, Graduate Student, Microbiology, University of Pennsylvania, Philadelphia

Ammiel Schwartz, Medical Student, New York University, New York City

George C. Sponsler, Graduate Student, Princeton University

Niccolo Visconti, M.D., Milan, Italy

Marguerite Vogt, M.D., Genetics, Institute for Brain Research, Neustadt, Germany

In connection with the course, a series of lectures was given by students, summer research workers, and visitors to the Laboratory. The speakers and their topics are listed below:

S. S. Cohen—Chemical studies on infected bacteria.

L. A. Manson—Metabolism of desoxyribose nucleosides.

M. H. Adams—Stability of viruses.

S. Granick—Speculations in biosynthesis.

N. Visconti—Kinetics of the lethal action of nitrogen mustard on bacteria and phage.

G. T. Barry—Counter current distribution.

R. Loevinger—Properties of ionizing radiations.

R. D. Hotchkiss—Transformation of pneumococcus.

M. Delbruck—Multiplicity reactivation and photoreactivation.

G. Stent—Adsorption cofactor requirements of phage T4.

COURSE ON BACTERIAL GENETICS

July 17-August 5, 1950

Instructors: Evelyn M. Witkin, Vernon Bryson, and M. Demerec, Carnegie Institution and Biological Laboratory.

Assistants: Frank L. Kennedy and Eileen Yongen, Carnegie Institution and Biological Laboratory.

A course on selected methods in bacterial genetics was offered this year for the first time. One of a very few of its kind given throughout the country, the course was organized and supervised by Drs. Witkin, Bryson, and Demerec, and was open to twelve advanced graduate and postdoctoral students. The purpose of this course was to acquaint students with some of the techniques used in the study of bacterial heredity, and to analyze critically some of the recent work done in this field.

Laboratory sessions were held six mornings a week for the three-week duration of the course, and the afternoons were devoted to lectures and informal discussion. Among the experiments conducted were the following: techniques of isolation and identification of mutants resistant to bacteriophages, radiation, or antibiotics, and of mutants requiring nutritional supplements for growth; techniques of measurement of spontaneous mutation rates by several methods; induction of mutations in bacteria by radiation and by chemical agents; and crossing of bacterial strains in which a sexual process can be demonstrated.

Many of the prominent summer visitors to the Laboratory cooperated by offering demonstrations and lectures that added greatly to the scope of the course. Dr. Rollin D. Hotchkiss, of the Rockefeller Institute for Medical Research, demonstrated techniques used in the study of type transformations in pneumococcus. A demonstration of cytological methods in bacteriology was given by Professor H. L. Chance, of the University of Oklahoma. Special lectures were given by Drs. M. Delbruck, of the California Institute of Technology; N. Visconti, of the University of Milan, Italy; J. S. Gots, of the University of Pennsylvania; M. Lieb, of Columbia University; F. M. Burnet, of the Walter and Eliza Hall Institute of Medical Research, Melbourne, Australia; and Mark H. Adams, of New York University.

The twelve students, selected from among a large number of applicants, were varied in background and interests, and brought to the course a wide range of viewpoints. The students were:

Dougles C. Appleby, Syracuse University, Syracuse, N. Y.

Robert G. Coon, Carnegie Institution, Cold Spring Harbor

Alan B. Cooper, Wesleyan University, Middletown, Conn. .

Alan Garen, University of Colorado, Denver

Joseph S. Gots, University of Pennsylvania School of Medicine, Philadelphia

N. Grossowicz, University of Jerusalem, Palestine

Gordon Lark, University of Chicago

Norman E. Melechen, University of Pennsylvania

Virginia Lee Speer, College of William and Mary, Norfolk, Va.

C. Tal, Rockefeller Institute for Medical Research, New York City

Niccolo Visconti, Milan, Italy

Maguerite Vogt, Institute for Brain Research, Neustadt, Germany

The course will be offered each summer as part of the regular program of the Laboratory.

PHAGE MEETING

August 21-22, 1950

Advantage was taken of the coincidence that several phage workers were going to be in or near Cold Spring Harbor towards the end of August, to call a phage meeting. It was attended by about 35 people, approximately half of whom came from some distance especially for this meeting. Morning, afternoon, and evening sessions were held on both days.

A. Garen presented results from T. T. Puck's laboratory, concerned with the mechanism of adsorption of phage to bacteria. Cations play a decisiv role in the process, and this role has been analyzed quantitatively and, in great detail. A remarkable similarity has been found between the conditions favoring adsorption of phage to bacteria and those favoring adsorption to glass. This similarity extends even to the role of the adsorption cofactor L-tryptophane. It appears that desorption of phage from the bacteria can be effected under conditions which prevent processes that normally follow adsorption from taking place.

J. D. Watson briefly discussed "Lysis from Without" (see "Viruses 1950"). z

M. H. Adams and J. D. Watson described two mutants of T5 and demonstrated that each of these mutants can give mixed infection in combination with wild-type T5. Adams in addition described experiments involving a phage that is serologically related to T5. A host susceptible to both T5 and the new phage has been found. Mixed infection of the common host with the two phages is possible, and genetic experiments on this pair are under way.

A. D. Hershey mentioned one experiment in which two mutants of T1 had been crossed and found to give recombinants.

M. Delbruck reported on experiments of R. Dulbecco on the interrelations between photoreactivation and multiplicity reactivation in T2. These experiments led to the suggestion that T2 consists of a special part and a genetic part. The special part is functional in the early phases of the infectious process. It is gradually discarded by the infecting particles. The genetic part then multiplies. After multiplication, new special parts are generated by the genetic parts. Many of the peculiarities in the radiobiology of T2, found by Dulbecco, can be explained qualitatively by this notion. Also, recent findings of S. Benzer, on the ultraviolet sensitivity of infected bacteria, fit this scheme.

G. Bertani presented his findings with the lysogenic coli strain of Lisbonne and Carrere. A convenient method for assaying the amount of free phage in the lysogenic culture was described. The method consists in first obtaining a streptomycin-resistant mutant of the indicator strain, and then plating a lysogenic culture, which is streptomycin-sensitive, on the indica-

tor strain in the presence of streptomycin. It has been shown that in this strain the phages are liberated in bursts. The strain liberates at least two types of phages, and whenever a bacterium produces a burst it is of either one or the other type. These findings very nicely parallel those recently reported by A. Lwoff for Bacillus megatherium.

A. H. Doermann reported on a mutable strain of T4. Four loci can be distinguished, and genetic crosses are under way to clarify their mutual relations. Doermann also reported a precision experiment on the proportion relations. Doermann also reported a precision experiment on the proportion of recombinants obtainable (by premature lysis) from a mixedly infected bacterium at various times during the latent period. It is found that recombinants are present from the very earliest stages at which infective particles are present, but that the proportion of recombinants increases by a factor of about 1.5 between this earliest stage and the end of the latent period.

S. S. Cohen reported on studies in carbohydrate metabolism which have clarified the origin of ribose and desoxyribose phosphates. These studies permit the formulation of a simple scheme accounting for the principal metabolic changes that occur in bacteria upon infection. Cohen also summarized experiments on chemical changes occurring in bacteria infected with ultraviolet-treated T2r+. The experiments demonstrate that UV treatment results in an extended delay in the onset of DNA synthesis. However, the synthesis of UV-absorbing materials, presumably constituents destined to be incorporated in DNA, is not inhibited in such systems.

A. F. Graham reported on experiments with phages labeled by P^{32}. The fate of the phosphorus of the infecting particles was studied, and a most remarkable difference was found depending on whether the particle was attacking an uninfected bacterium or a bacterium which had been infected by another particle at least two minutes earlier. If the bacterium had not been previously infected, very little of the label of the infecting particle appeared as inorganic phosphorus in the medium, and 20% of the label reappeared in the phage yield. In contrast, if the bacterium had been previously infected, then 50% of the label of the secondarily infecting particle appeared in the medium within a few minutes.

A. Novick described the "chemostat" designed by Novick and Szilard, in which bacteria can be grown in a constant environment for an indefinite length of time and at any desired growth rate. Novick also presented some preliminary data on several mutation rates and their dependence on the growth rate, and on temperature.

T. F. Anderson described a new and ingenious method for avoiding many of the drying artifacts of electron microscopy. Specimens are first transferred from the aqueous solution into liquid carbon dioxide through a series of mutually miscible intermediate solvents. The liquid carbon dioxide is then removed via a path that circles the critical point in the pressure-volume diagram of carbon dioxide. In this manner, specimens are obtained

which are not distorted by any of the surface-tension forces usually associated with the drying process. Anderson showed some beautiful stereoscopic pictures of preparations obtained in this manner, particularly pictures of phage T2 adsorbed on bacteria. It was quite evident from these preparations that phage T2 is adsorbed to the bacteria tailfirst.

S. E. Luria presented new data on the distribution of the number of spontaneous mutants in the yields from individual bacteria. These results indicated that the mutants are distributed clonally and that they multiply by binary fission. Luria also presented recent data on the comparison of particle counts, obtained with the electron microscope, and plaque counts obtained from the same purified phage preparations. The particle count and the plaque count in all cases agreed within a factor of two or better.

A. D. Hershey summarized the proceedings of the meeting.

A square dance was held during the later part of the first evening, and a beer-and-pizza party concluded the proceedings of the second night. In the court of this last session M. Delbruck was tried in the Yokel Court of Long Island on charges of spitting into his cultures. Judge Loevinger presided. After numerous witnesses had been called and cross-examined, the jury returned a verdict of "guilty", and the defendant was sentenced to twenty years of hard labor in the California Penitentiary of Technology.

NATURE STUDY COURSE

July 5 — August 8, 1950

Dr. and Mrs. J. Southgate Y. Hoyt, Laboratory of Ornithology, Cornell University. Assisted by Martin Pierce and Susie Mayr

With the 1950 season, the Nature Study course started on its second decade. It was in 1940 that the work was begun, with Dr. Hoyt handling it alone. In the ten years, it has grown and developed so that it takes the full time, for the five-week period, of both Dr. and Mrs. Hoyt and two assistants.

The course started this year on July 5th, and again was conducted with the following aims: (1) that the students learn to see the many interesting plants and animals that occur abundantly about them all the time; (2) that each student learn how to make careful observations of the things he sees, and learn how to answer his own questions about his observations; (3) that each student learn that no one is too young or too much of an amateur to make contributions to knowledge by careful and accurate study of objects and phenomena observed.

Classes were conducted very informally, beginning with a discussion of plants, animals, and objects brought in by the children themselves, or of new acquisitions in the laboratory since they last met as a class. Then the group went outdoors to visit various habitats, to collect anything that interested them and bring it back to the laboratory for more careful study. The Biological Laboratory is ideally situated for such studies, for within short walking distance one may visit large fresh water ponds, swift heavily shaded streams, hot slow streams, brackish water habitats, heavy forest areas of deep shade and moist floor, open fields, sandy sea beach, tide pools, salt swamps, and mud flats. The ecological relationships of the life found in these localities was stressed at all times. Wawepex Laboratory was headquarters for the work, and specimens were brought there to be studied and added to the collection of living forms kept on hand during the course. The laboratory was always open, and each student was encouraged to return at any time, to work, to ask questions, or to bring things he found at home or on his walks.

Twenty-four Juniors, aged 6 to 8 years, met Monday and Wednesday mornings from 9:00 to 11:00. This group made three trips to the beach on the sand spit, and found many interesting forms of life, at both low and high tide. Through the courtesy of the staff members of the New York State Fish Hatchery, this and the other classes had a chance to see the methods of raising trout to stock our streams, and learned the need for conservation of our natural resources. Other field trips were taken to nearby lakes, woods, and fields.

Twenty-two Intermediates, aged 9 to 11 years, met Tuesday and Thursday mornings from 9:00 to 11:00. This group also visited the beach and lakes, studied the life in a nearby warm pond in contrast to that of a cold lake, and devoted an indoor period to making exhibits of leaves they had collected and identified. Both groups had the opportunity, one rainy

day, to see films of unusual animals and birds, taken by Dr. Hoyt in the London Zoos.

The three Seniors, aged 12 to 14 years, met two afternoons a week. These students collected and maintained pond cultures, studying them microscopically from time to time to see the changes that occurred in standing water. A trip was taken one afternoon to the Roosevelt Bird Sanctuary at Oyster Bay, where the Director, Mr. James Callaghan, demonstrated and explained purposes of the exhibits in the Museum, and took the youngsters along the Nature Trail.

All classes helped maintain aquaria and terraria in the laboratory, and added animals and plants to them when possible. A number of the larger moths were found and brought in, and as some of these laid eggs in the laboratory, there was an opportunity to see a portion of the life cycle of such an insect; some of the eggs hatched before the course ended. The students also learned how to mount moths and butterflies for their own collections. Shell and rack collections were also started.

The classes ended on August 8th, with a public exhibition of the activities and of the materials collected. The students were encouraged and helped to prepare demonstrations of their particular interests and to arrange the material in an attractive manner for inspection by their parents and friends. At two o'clock the laboratory was opened for the exhibit, and all visitors were impressed with the wealth of interesting material, collected within a short distance of Wawepex or the students' homes. At three o'clock, a Kodachrome movie was shown, in Blackford Hall, of the life history of the Monarch butterfly, as well as a series of Kodachrome slides showing native animals and plants, all taken by Dr. Hoyt. Ice cream and cake were served by the laboratory to all present.

The following children attended the classes.

Abramson, Barbara
Adams, Gay
Avery, Bramman
Baty, Carl
Belmont, August
Berry, Roger
Bryson, Stephen
Buckley, Sarah
Cleaveland, Edwards
Cleaveland, Peter
Flessel, John
Gots, Ronnie
Granick, Lee
Granick, Donna
Griffiths, Clare
Griffiths, Kathleen
Grossowicz, Yoram
Hewitt, Lindsay
Holske, Robin
Hotchkiss, Cynthia
Hotchkiss, Paul
Laverne, Daniel
Laverne, Jerry
Lyon, Alexander
Lyon, Richard
Makanna, Martha
Makanna, Philip
Nields, John
Nields, Elizabeth
Noyes, Sandy
Page, Jane
Parsons, Llewelyn
Radsch, Bobby
Radsch, Chris
Radsch, Dick
Rippere, Kenneth
Rippere, Robert
Rutherford, John
Saarinen, Susie
Schneider, Ann
Schneider, Kate
Shemin, Louise
Sheshunoff, Alex
Smaridge, Alex
Smaridge, Courtenay
Tallman, Phoebe
Tittler, Robert
Vacquier, Victor
Watkins, John

COLD SPRING HARBOR SYMPOSIA ON QUANTITATIVE BIOLOGY

Current volume: XV. Origin and Evolution of Man. 425 + xii quarto pages, 53 figures. 1950. Table of contents is listed below:

POPULATION AS A UNIT OF STUDY

STRANDSKOV, H. H.—The genetics of human populations.
BUZZATI-TRAVERSO, A.—Genetic structure of natural populations and inter-breeding units in the human species.
THIEME, F. P.—Problems and methods of population surveys.
BATES, M.—Concluding remarks of the chairman.

ORIGIN OF THE HUMAN STOCK

SCHULTZ, A. H.—The specializations of man and his place among the catarrhine primates.
SIMPSON, G. G.—Some principles of historical biology bearing on human origins.
WASHBURN, S. L.—The analysis of primate evolution with particular reference to the origin of man.
HOWELLS, W. W.—Concluding remarks of the chairman.

CLASSIFICATION OF FOSSIL MEN

McCOWN, T. D.—The genus Palaeonanthropus and the problem of superspecific differentiation among the Hominidae.
STEWART, T. D.—The problem of the earliest claimed representatives of Homo sapiens.
MAYR, E.—Taxonomic categories in fossil hominids.
KROGMAN, W. M.—Concluding remarks of the chairman.

GENETIC ANALYSIS OF RACIAL TRAITS (I)

BOOK, J. A.—Clinical and genetical entities in human populations, with some remarks on schizophrenia, manic-depressive psychosis and mental deficiency in a North Swedish population.
KEMP, T.—The frequency of diseases affected by heredity in Denmark.
NEEL, J. V.—The population genetics of two inherited blood dyscrasias in man.
SNYDER, L. H.—Concluding remarks of the chairman.

GENETIC ANALYSIS OF RACIAL TRAITS (II)

LAUGHLIN, W. S.—Blood groups, morphology and population size of the Eskimos.
SPUHLER, J. N.—Genetics of three normal morphological variations; patterns of superficial veins of the anterior thorax, peroneus tertius muscle and number of vallate papillae.
LASKER, G. W.—Genetic analysis of racial traits of teeth.
REED, S. C.—Concluding remarks of the chairman.

GENETIC ANALYSIS OF RACIAL TRAITS (III)

BOYD, W. C.—Three general types of racial characteristics.
RACE, R. R.—The eight blood group systems and their inheritance.
MOURANT, A. E.—The blood groups of the peoples of the Mediterranean area.
OLIVER, C. P.—Concluding remarks of the chairman.

RACE CONCEPT AND HUMAN RACES

COON, C. S.—Human races in relation to environment and culture, with special reference to the influence of culture upon genetic changes in human populations.
BIRDSELL, J. B.—Some implications of the genetical concept of race in terms of spatial analysis.
MONTAGU, M. F. A.—A consideration of the concept of race.
LUNDMAN, B.—Anthropological maps of the Nordic countries.
ANGEL, J. L.—Population size and microevolution in Greece.
DUNN, L. C.—Concluding remarks of the chairman.

CONSTITUTION

GARN, S. M.—The constitutional modification of Mendelian traits in man.
SELTZER, C. C.—Constitutional aspects of juvenile delinquency.
SHELDON, W. H.—The somatotype, the morphophenotype, and the morphogenotype.
HOOTON, E. A.—Concluding remarks of the chairman.

PERSPECTIVES OF FUTURE RESEARCH

DOBZHANSKY, T.—Human diversity and adaptation.
KLUCKHOHN, C., and GRIFFITH, C.—Population genetics and social anthropology.
STERN, C.—Concluding remarks of the chairman.

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 Relations 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.
Vol. XIII (1948) Biological Applications of Tracer Elements, 220 pp.
Vol. XIV (1949) Amino Acides and Proteins, 217 pp.

* Out of print.

LABORATORY STAFF

Albers, Carl—Engineer
* Bass, Emma—Maid
* Brandt, Jane—Research Assistant
* Branton, Geneva—Cook
Bruno, Dominic—Gardener
Bryson, Vernon—Geneticist
* Byrne, Patrick—Laborer
Cosillo, Gloria—Technical Assistant
Cuneo, Helen—Research Assistant
Demerec, M.—Director
* DeRosa, Jerry—Laborer
Dorsey, Henry—Laborer
Farrington, Margaret—Technical Assistant
* Fedoroff, Alex—Technical Assistant
Fickert, Kurt—Carpenter
Forgione, Louis—Research Assistant
Franzese, Eleanor—Clerical Assistant
* Gardner, Della—Cook
Gardner, Henry—Technical Assistant
Hershey, Harriet D.—Research Assistant
* Hoyt, J. Southgate Y.—Nature Study Course Instructor
Hsie, Jen-yah—Bacteriologist
Kaplan, Selma—Research Assistant
King, James C.—Research Associate
Klem, Dorothy V.—Secretary
Lowell, Francis—Superintendent of Grounds
Madden, Carol V.—Research Assistant
* Mayr, Christa—Technical Assistant
* Mirel, Emanuel—Research Assistant
Monsees, Howard—Research Assistant
* Payne, Ruth—Cook
† Plunkett, Geoffrey, Jr.—Research Assistant
* Prout, Timothy—Research Assistant
† Prytz, Bo—Chemist
† Rae, William S.—Superintendent of Grounds
† Raymond, Constance—Research Assistant
Reddy, William—Laborer
Rosenblum, Eugene—Bacteriologist
* Schultz, Peter—Technical Assistant
* Schultz, Judith—Technical Assistant
* Streisinger, George—Research Assistant
Wallace, Bruce—Geneticist
* Warren, Katherine B.—Editor of Symposium manuscripts
† Wright, Theodore—Research Assistant
† Yongen, Eileen—Research Assistant
* Yongen, Lillian—Dining Hall Manager

* Summer and Temporary
† Resigned during the year.

SUMMER RESEARCH INVESTIGATORS

Abramson, Harold A.—Cold Spring Harbor, N. Y.
Adams, Mark H.—New York University College of Medicine, New York, N. Y.
Atchley, William A.—The Rockefeller Institute for Medical Research, New York, N. Y.
Bernheimer, Alan W.—New York University College of Medicine, New York, N. Y.
Burnet, F. M.—The Walter and Eliza Hall Institute of Medical Research, Melbourne, Australia.
Caspari, Ernst—Wesleyan University, Middletown, Connecticut.
Collins, Nancy—New York University College of Medicine, New York, N. Y.
Colowick, Sydney—John Hopkins University, Baltimore, Maryland.
Delbruck, Max—California Institute of Technology, Pasadena, California.
Diaz Collazo, Ana M.—University of Puerto Rico, Rio Piedras, Puerto Rico.
Fano, Ugo—National Bureau of Standards, Washington, D. C.
Granick, S.—The Rockefeller Institute for Medical Research, New York, N. Y.
Hotchkiss, Rollin D.—The Rockefeller Institute for Medical Research, New York, N. Y.
Lark, Gordon—New York University College of Medicine, New York, N. Y.
Mayr, Ernst—The American Museum of Natural History, New York, N. Y.
Miller, Elizabeth—The Rockefeller Institute for Medical Research, New York, N. Y
Racker, Ephraim—New York University College of Medicine, New York, N. Y.
Ratner, Sarah—New York University College of Medicine, New York, N. Y.
Sansome, Eva R.—University College, Ibadan, Nigeria.
Sansome, F. F.—University College, Ibadan, Nigeria.
Schultz, Jack—The Institute for Cancer Research, Philadelphia, Pa.
Shemin, David—College of Physicians & Surgeons, Columbia University, New York, N. Y.
Stekol, J. A.—The Institute for Cancer Research, Philadelphia, Pa.
Swanstrom, Maryda L.—New York University College of Medicine, New York, N. Y.
Schaeffer, Morris—U. S. Public Health Service, Communicable Disease Center, Montgomery, Alabama.
Tittler, Irving—Brooklyn College, Brooklyn, N. Y.
Vinogradoff, D.—National Bureau of Standards, Washington, D. C.
Visconti di Modrone, N.—Politecnico di Milano, Milan, Italy.
Vogt, Marguerite—Institut fur Hirnforschung, Neustadt, Germany.
Watson, James—California Institute of Technology, Pasadena, Calif.

REPORT OF THE SECRETARY

The 59th meeting of the Board of Directors of the Association was held on January 23, 1950, at the Broad Street Club in New York City, with fifteen members present and Dr. Robert Cushman Murphy presiding. The minutes of the last meeting were accepted as distributed; and the names of 208 contributors during 1949 were presented by the Secretary for ratification as Sustaining Members for the year 1950. The report of the Director of the Laboratory dealt first with the need for new buildings and improvements to old buildings, a program that could be carried out at an estimated cost of $300,000. He then reported on the program of the various research groups during the year 1949, describing one or two of the most interesting findings. Plans for the Symposium and the summer program were outlined, and the tentative program of the Symposium circulated. The report of the Treasurer was distributed and accepted; and the proposed budget for 1950, presented by the Laboratory Director, was unanimously approved. Approval was also voted for the purchase of a trlck and for the establishment of a Research Reserve of $3000. President Murphy, in accordance with a motion passed at the 1949 Annual Meeting, announced the appointment of a Nominating Committee, consisting of Dr. Haskins, as chairman, Mrs. Franklin and Mr. Morris.

A luncheon meeting of the Executive Committee was held on April 4, at the Harvard Club in New York City, to discuss a proposal that the present coordinated operation of the Biological Laboratory and the Department of Genetics be recognized by a more formal agreement between the Association and the Carnegie Institution of Washington. Dr. V. Bush, and Mr. Paul A. Scherer, President and Executive Officer of the Carnegie Institution, and Dr. M. Demerec, Director of both the laboratories, met with the Committee. Dr. Bush reviewed the past and present relations between the two laboratories; and Mr. Page discussed the situation from the standpoint of the Association. From the general discussion that followed it was evident that the Committee was unanimous in approving the suggestion to proceed at once with the formulation of an agreement, patterned after that adopted for the operation of the Mt. Wilson and Palomar Observatories. Dr. Bush agreed to submit to the Directors of the Association a draft of such a plan for their consideration.

The 27th Annual Meeting of the Association, held at Blackford Hall, Cold Spring Harbor, on July 25, 1950, was called to order by Vice President Arthur W. Page. The Secretary reviewed the chief acts of the Association during the past year, and this report was voted accepted. Since copies of the Annual Report were available in time for the meeting, the Laboratory Director gave only a brief report of the progress of research. He mentioned the recent very successful Symposium, and the two advanced courses offered by the Laboratory this summer. On the subject of the outstanding needs of the Laboratory, the Director reported that in March the Carnegie Corporation of New York had approved a grant of $100,000 for an auditorium building, to be used jointly by the Department of Genetics and the Biological Laboratory, and that in June the Laboratory had received from the Rockefeller Foundation a grant of $45,000 for im-

provements to present buildings and grounds. The Director's report was concluded with a discussion of the use of the special funds available for research, which had amounted to about $80,000 in the past year. These funds, fully adequate to support the research projects, cannot be used for the running expenses of the Laboratory, which must be met through the contributions of the membership. This report was approved. The Treasurer's report, as published in the Annual Report, was accepted in full. The following members were named by the Nominating Commitee and elected or re-elected to the Board of Directors to serve until 1954: Mrs. Walter H. Page, Mr. Amyas Ames, Dr. Robert Chambers, Dr. George W. Corner, Dr. Th. Dobzhansky, and Dr. Ernst Mayr.

The 60th meeting of the Board of Directors was held on July 25, 1950, at the close of the Annual Meeting of the Association. The minutes of the last meetings of the Board and of the Executive Committee were approved. Mrs. Franklin reported that the Women's Committee had been reorganized, and had recently been entertained at a tea, where an account of work at the Laboratory was given by Dr. Evelyn M. Witkin and plans for an Open House in September were discussed. The Executive Committee as constituted in 1950 was unanimously re-elected for the following year. It was moved and unanimously voted that a Resolution of Corporation be executed, authorizing temporary borrowing, in amount not exceeding $20,000, to meet emergencies resulting from the delayed payments from grants..

E. Carleton MacDowell, Secretary

LONG ISLAND BIOLOGICAL ASSOCIATION
FINANCIAL STATEMENT

MAIN AND COMPANY
Certified Public Accountants
New York, U.S.A.

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, 1951. 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 schedule, together with the notes thereon, present fairly the position of the Long Island Biological Association at April 30, 1951 and the results of its operations for the year ended on that date.

Main and Company
Certified Public Accountants

New York, N. Y.
July 3, 1951.

LONG ISLAND BIOLOGICAL ASSOCIATION

BALANCE SHEET

April 30, 1951

ASSETS

General and Endowment Fund			
Cash:			
In banks	$ 52,089.57		
On hand	100.00	$ 52,189.57	
Investments (market value $18,388.69)		17,564.06	
Accounts Receivable:			
National Tuberculosis Association	$ 608.93		
United States Department of the Army	8,391.89		
Miscellaneous	1,297.38	10,298.20	
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	$378,321.68
Special Funds			
Cash in bank		$ 522.12	
Investments (market value $15,301.58)		15,710.00	16,232.12
Total			$394,553.80

NOTE: In accordance with the Association's established practice, the above balance sheet does not include the inventory at April 30, 1951 of the published volumes of the Association's yearly Symposia of Quantitative Biology, nor has any depreciation or amortization on buildings and equipment been recorded on the Association's records. Additions and improvements to buildings and equipment have been charged against current operations in conformity with the Association's usual practice.

LIABILITIES AND NET WORTH

General and Endowment Fund			
Liabilities:			
Accounts payable		$ 7,386.66	
Special grants:			
Josiah Macy, Jr. Foundation	$ 789.17		
The Jane Coffin Childs Memorial Fund for Medical Research	405.27		
Polio Basic Research Fund of Long Island	540.09		
Rockefeller Foundation Grant	13,324.68		
United States Atomic Energy Commission	11,935.87		
Viking Fund	616.12	27,611.20	
Total Liabilities		$ 34,997.86	
Reserve for scientific research		3,000.00	
Endowment Fund:			
Dr. William J. Matheson Bequest		20,000.00	
Net Worth		320,323.82	$378,321.68
Special Funds			
Blackford Memorial Fund:			
Principal		$ 5,000.00	
Charles Benedict Davenport Memorial Fund:			
Principal	$ 4,934.75		
Unexpended income	412.75	5,347.50	
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	$ 2,247.48		
Unexpended income	37.52	2,285.00	16,232.12
Total			$394,553.80

STATEMENT OF NET WORTH

For the Year Ended April 30, 1951

Balance, May 1, 1950	$319,224.33
Add:	
Excess of income over expense for the year ended April 30, 1951	1,099.49
Balance, April 30, 1951	$320,323.82

STATEMENT OF INCOME AND EXPENSE

For the Year Ended April 30, 1951

Income:			
Contributions:			
Dues and contributions	$ 5,548.50		
Carnegie Corporation (grant for annual Symposia)	6,000.00		
Wawepex Society	1,650.00		
John D. Jones Scholarship	250.00	$13,448.50	
Symposia:			
Book Sales	$11,924.10		
Registration fees	21.02	11,945.12	
Dining hall		9,447.34	
Rooms and apartments		8,121.50	
Research fees		6,265.63	
Interest and dividends on investments		895.29	
Other Income:			
Summer course tuition	$ 1,220.00		
Nature course study	31.30		
Annual distribution from Walter B. James Fund	70.00	1,321.30	
Total Income			$51,444.68

Expense:			
Symposia:			
Publication of annual Symposia on Quantitative Biology	$ 6,858.22		
Expense of participants and lecturers	7,591.10	$14,449.32	
Dining Hall		11,565.02	
Rooms and apartments		1,884.84	
Research expenses		1,841.46	
Summer course expense		463.25	
Loss on sale of securities		8.91	
Bad debt		400.00	
Distribution of John D. Jones Scholarship		150.00	
Buildings and grounds maintenance:			
Salaries	$ 7,031.26		
Materials and supplies	2,317.13		
Heat, light and water	2,281.67	11,630.06	
General and Administrative:			
Salaries	$ 4,245.95		
Insurance	1,393.21		
Printing and stationery	1,102.25		
Telephone, telegraph and postage	427.55		
Miscellaneous	783.37	7,952.33	
Total expense			50,345.19
Excess of income over expense			$ 1,099.49

STATEMENT OF SPECIAL GRANTS

For the Year Ended April 30, 1951

	Balances May 1, 1950		Transactions May 1, 1950 to April 30, 1951			Balance April 30, 1951	
From Whom Received	Due to Association (Accounts Receivable)	Unexpended Balance of Grant	Amounts Received	Expenditures Charged Against Grant	Income to Association Charged Against Grant	Due to Association (Accounts Receivable)	Unexpended Balance of Grant
Josiah Macy, Jr. Foundation		$ 647.66	$ 650.00	$ 408.49	$ 100.00		$ 789.17
The Jane Coffin Childs Memorial Fund for Medical Research		1,536.83		881.56	250.00		405.27
National Tuberculosis Association	$ 171.86		5,115.58	5,139.49	413.16	$ 608.93	
Polio Basic Research Fund of Long Island		1,793.00		1,252.91			540.09
Rockefeller Foundation Grant			45,000.00	31,675.32			13,324.68
United States Atomic Energy Commission		13,206.67	32,105.00	30,963.48	2,412.32		11,935.87
United States Dept. of the Army	13,969.88		30,475.62	22,257.48 883.88	2,640.15	8,391.89	616.12
Viking Fund			1,500.00				
Totals	**$14,141.74**	**$17,184.16**	**$114,846.20**	**$93,462.61**	**$5,815.63**	**$9,000.82**	**$27,611.20**