

(Papers read before the Institute but not published in the Proceedings.)

INTERPRETATION OF THE TUBERCULIN TEST. C. B. STEWART. (Read November 16, 1953). A brief review was given of the method of preparation of tuberculin and the methods of administration of the tuberculin test. Data were presented on the tuberculin reactions of 1292 students in the Halifax schools of nursing who were in training between 1947 and 1953.

The proportion of reactors to the Vollmer Patch test of ten tuberculin units accurately reflected the infection rate in the Nova Scotia population. There was a slight downward trend in the proportion of reactors from 1947 to 1953 paralleling a decrease in this disease throughout the province. There was also a definite increase in the proportion of reactors with age. On the other hand, the stronger test of 250 tuberculin units (2nd strength purified protein derivative of tuberculin) showed wide and apparently erratic fluctuations from year to year in the proportion of reactors and was not correlated with increasing age.

It was also demonstrated that positive reactions to the dose of ten tuberculin units was very stable, with only 0.5 per cent reverting to negative each year during a four year follow up. On the other hand 23.3% of the reactors to 250 tuberculin units were negative one year later. The incidence of tuberculosis was also higher among reactors to 250 tuberculin units than among the reactors to ten tuberculin units.

It was concluded that a high dose tuberculin test such as second strength purified protein derivative produces a large proportion of non-specific reactions and should not be used as an index of a previous tuberculous infection.

THE RELATION OF PROTEIN TO THE CHYLOMICRON. C. B. WELD. (Read November 16, 1953) The lipid component of the chylomicron of an alimentary lipemia has been reported by various authors to be chiefly neutral fat. However, electrophoresis and ultracentrifuge studies place the chylomicrons in a lipoprotein fraction, and I have previously reported evidence indicating that the chylomicron must have a protein moiety. This has been studied with the following results.

By overlaying a lipemic serum with saline (pH 7.4) and centrifuging at 8,000-10,000 RPM a relatively pure suspension of chylomicrons is obtained. If this is now removed, made up with saline to the original volume, and centrifuged at 10,000 RPM the chylomicrons rise to form an opaque zone with a clear supernatant fluid. With care, the chylomicron layer can be pipetted off and transferred to a fresh volume of saline. It was hoped that after several such washings the protein of the suspending fluid would be completely removed, leaving only that protein that was bound as lipo-protein in the chylomicron.

It is found that, after 3 washings, the lipid content of the chylomicron layer has only been cut in half while the protein content has been reduced to about 2% of its former value. After four washings there is danger of the chylomicrons coalescing and forming a pellicle and thus being no longer chylomicrons. At this unstable period the chylomicron layer usually has a protein content of less than 0.002% and the proportion of protein to lipid is about 1% or less.

It is therefore concluded that in a lipemia the chylomicrons consist of lipid globules kept dispersed by an interfacial protein film of molecular dimensions and that very little protein, if any, is bound to the lipid as a lipo-protein complex.

AN EPIDEMIC OF ACUTE NEPHRITIS R. W. REED. (Read November 16, 1953.) Twenty-two cases of acute nephritis occurred in a localized rural area of Nova Scotia between October 1951 and January 1952. School attendance records and family illness surveys for this period showed a high absentee rate and a high incidence of acute sore throat. Throat swabs from a large group of individuals were cultured and anti-streptolysin titrations done on as many cases and contacts as possible. An unusually high carrier rate of type 12 hemolytic streptococci was revealed.

The laboratory and clinical findings in this epidemic are reported and discussed.

DIKARYONS IN CHLAMYDOMONAS MOEWUSII. RALPH A. LEWIN. (Read December 14, 1953.) *Chlamydomonas moewusii* is heterothallic but isogamous. Pairing gametes become associated first by their flagella, and then by a protoplasmic bridge between the papillae. *Vis-a-vis* pairs normally are actively motile for some hours; they remain in this stage for days if kept in darkness, since light appears essential for plasmogamy. The *plus* partner is solely responsible for the propulsion of the pair. Pairs in which the *plus* cell is genetically paralyzed are always initially immotile; whereas, in pairs between wild-type *plus* and paralyzed *minus* cells, motility is normal.

Certain U-V induced mutants have been found in which paralyzed *plus* gametes, mated with *minus* cells of wild-type or of different mutant stocks, recover motility after two to forty-eight hours (depending on the strain tested); and it can be shown by the use of markers that this is due to recovery of motility by the *plus* gamete. It is therefore concluded that, in these morphologically normal mutants, paralysis is a consequence of a genetic deficiency in certain as yet unidentified motility factors, which can be transmitted from one gamete to another. Since filtrates of wild-type cells do not induce recovery of paralyzed cells, transmission apparently takes place through the protoplasmic bridge. In this way, two nuclei of dissimilar genotype are capable of interacting within a single protoplasmic continuum, as in fungal heterokaryons, and suitable combinations are capable of producing phenocopies of wild-type.

There is no indication that the postulated motility factors are self-reduplicating: hybrid zygosporangia (mutant x wild type) segregate normally 2:2. In the case of one mutant, the paralysis gene is very closely linked with mating-type, no recombination having been obtained from over fifty zygotes analyzed.

SOME LOCAL TEMPERATURE VARIATIONS IN HALIFAX. REID V. DEXTER. (Read December 19, 1953.) A comparison of daily maximum temperatures at 14 Mayfield Avenue, with the official downtown maxima is presented for the months of May, June, July, August and September, 1953. Differences as high as 11°F. are shown. Conditions of wind and sunshine conducive to large differences are pointed out, but it is shown that no ready rule of thumb can be evolved to make possible the computation of the Mayfield Avenue temperature from the downtown temperature and the general meteorological conditions.

The results of daytime temperature measurements made during four trips around the city in an automobile equipped with a portable thermometer are tabulated. Temperature variations from one part of the city to another at approximately the same time are as high as 14°F.

These variations are related qualitatively to the wind and the topography of the city.

AN APPRAISAL OF CANADIAN NUTRITURE. E. GORDON YOUNG. (Read December 14, 1953.) From a review of data available at present it may be concluded that Canadians have available adequate quantities of all essential nutrients if consumed according to individual requirements. School children across the country show an appreciable percentage who are underweight and exhibit signs of past and present deficiencies of vitamin D and calcium, and possibly of riboflavin. Local groups of adults show general undernutrition and deficiencies of vitamins A, B₂, C and niacin. Local groups of both children and adults are not eating a well balanced diet. A condition of mild anemia occurs in school children between the ages of five and fourteen years to the extent of probably less than five percent in most localities surveyed but that in certain sections of the country it may be much higher. This state of affairs also applies to about five per cent of young women in certain areas. Overweight in adults probably represents the commonest form of malnutrition and may be applicable to one fifth of the population over thirty years of age.

EVIDENCE FOR THE ROLE OF REDUCED SULPHUR COMPOUNDS IN SILICON UTILIZATION BY DIATOMS. JOYCE LEWIN. (Read December 14, 1953.) 1. Cells of the freshwater diatom *Navicula pelliculosa* may be grown in a mineral medium containing a low concentration of silicon. When transferred to a fresh silicate solution and incubated under non-growing conditions such depleted cells rapidly take up silicon from the medium.

2. The utilization of silicon is an aerobic process.

3. When depleted cells are washed with distilled water or saline, their ability to utilize silicon is impaired whereas respiration is unaffected.

4. The ability of washed cells to take up silicon can be partially restored with sulfate or ascorbic acid, and is completely restored by Na₂S, Na₂S₂O₃, glutathione, l-cysteine, dl-methionine, or ascorbic acid plus sulfate.

5. The sulfhydryl reagent, CdCl₂, inhibits silicon utilization of unwashed cells at concentrations which do not affect respiration. This inhibition similarly is reversed by glutathione or cysteine.

6. However, sodium iodoacetate and sodium arsenite inhibit respiration and silicon utilization at the same concentrations.

7. Sulfhydryl groups in the cell membrane may be involved in silicon uptake by diatoms.

EXPERIMENTAL NEPHRITIS DUE TO TYPE SPECIFIC STREPTOCOCCI. R. W. REED AND B. H. MATHESON. (Read January 11, 1954.) Several strains of type 12 streptococci recovered from throat cultures in widely separated outbreaks of acute glomerulonephritis have been used in these studies. Localized infection of rabbits with these strains has resulted in the appearance of hypertension, albuminuria and hematuria eighteen days following infection. Intravenous injection of filtrate from these cultures results in a similar picture eight days after the beginning of treatment. Animals infected locally with other serological types of streptococci and with unrelated bacteria did not develop hypertension or urinary abnormalities. The significance of the clinical and patho-

logical findings and their relation to acute glomerulonephritis in man is discussed.

VISCOSITY AND MOLECULAR WEIGHT OF DEGRADED CARRAGEENIN. C. R. MASSON AND G. W. CAINES. (Read January 11, 1954.) Viscosities of aqueous solutions of carrageenin, including thermally and photochemically degraded preparations, have been measured both in the presence and absence of added salts. Measurements were made using Ostwald and Stormer viscometers. Number-average molecular weights were determined osmotically.

The viscosity characteristics of these solutions are entirely similar to those of other natural and synthetic polyelectrolytes. In the absence of salts, the viscosity-concentration relationship may be expressed by the Fuoss equation. The results are interpreted in terms of the shape of the carrageenin molecule in solution.

The relationship between viscosity and molecular weight indicates that the molecules exist in solution as fairly rigid rods, even in the presence of salts.

VARIATION OF THE HAEMOGLOBIN LEVEL WITH AGE AND SEX. W. W. HAWKINS, E. SPECK and VERNA G. LEONARD. (Read January 11, 1954.) During the winter and spring of 1952-53 a survey of haemoglobin values was made in Halifax among 1308 male subjects 6 to 98 years of age, and 1424 female subjects 6 to 94 years of age.

Capillary blood was used. Pigment was determined by converting to cyanmethaemoglobin and reading the optical density in a Coleman spectrophotometer. This method was standardized against blood of which the haemoglobin concentration had been determined from the iron content and oxygen capacity.

Among children 6 to 14 years old the values increased from about 13 to about 14 Gm. % (Gm. per 100 ml. of blood) and there were essentially no differences between the sexes. The average value for both the boys and the girls was 13.5 Gm. %, with standard deviations of 1.00 and 0.96.

In girls between 14 and 20 years of age the haemoglobin values decreased slightly, reaching about 13 Gm. %. In boys of corresponding ages there was an increase to about 15 Gm. %. In both sexes these values were attained at about 20 years of age, and remained characteristic of the third decade of life. They were essentially the lowest and the highest shown respectively by the female and the male subjects of any age group.

Haemoglobin values in men between 20 and 60 years of age were typically 14.5 to 15.0 Gm. %, the higher values tending to occur among the younger men. After the fifth decade there were progressive and marked decreases to an average of 12.4 Gm. % in men between 80 and 90 years of age.

In women from 20 years of age onward the average haemoglobin values remained near 13 Gm. %.

Analysis of the figures obtained for the various age groups of both sexes showed standard deviations ranging from 0.69 to 1.73, with usual values of about 1.

THE EFFECTS OF VISIBLE RADIATION ON THE OVOTESTIS OF THE SLUG, *Deroceras Reticulatum* (MULLER). D. PELLUET AND N. E. HENDERSON. (Read February 8, 1954.) A brief summary is given of spermatogenesis in the hermaphroditic slug, *Deroceras reticulatum*.

The abnormal types of male germ cells which normally occur in this species are described. Light, up to an intensity of 1,500 mc., induced a rapid maturation of male germ cells (which resulted in a loss of organization within the follicles) following which senescent changes occurred within the gonad. Animals kept in total darkness indicated a premature senescence, so that some light is required in the normal environment of these nocturnal animals. Intensities of light beyond 1,500 mc. inhibited both nuclear and cytoplasmic division in spermatocyte cells, but not their differentiation. Thus, both primary and secondary spermatocytes matured directly, having omitted both meiotic divisions. The inhibition of cytoplasmic divisions was also manifested in the increase of binucleate spermatocytes and the large proportion of multinucleate spermatids. Temperature increases of only a few degrees above normal room temperature were also effective in producing similar responses, but these were overshadowed by changes comparable to those occurring in heat-sterility in vertebrates. The mechanism by which light and heat may act to produce these changes is discussed.

DIFFRACTION OF AN ACOUSTIC WAVE OBLIQUELY INCIDENT UPON A PLANE OBSTACLE. H. S. HEAPS. (Read February 8, 1954.) Some calculations have been undertaken to determine the variation of sound pressure in the region behind a rigid plane disc irradiated by a plane wave of sound. The dimensions of the disc are assumed to be comparable to the wavelength of the incident sound. Comparison is made with the case in which the plane of the disc is at right angles to the direction of the wave.

THE EXPANSION OF MONOMOLECULAR FILMS OF PROTEIN. J. GORDIN KAPLAN and M. J. FRASER. (Read February 8, 1954.) It has been known for some time that a monolayer of protein maintained at constant surface pressure at the air/water interface will tend to expand with time. An alternative of hypotheses is available to explain this phenomenon: 1. There is a progressively more complete unfolding of the protein molecules themselves as a result of the asymmetric field of force at the interface; 2. There is a progressive, irreversible adsorption at the interface of molecules which had escaped to the bulk phase at the time of spreading. Hypothesis 1 seems to be accepted at the present time, although Irving Langmuir suggested in 1938 that progressive migration to the surface of originally dissolved molecules might account for changes in area of protein films with pH. We have devised a very simple two-trough experiment to decide in favour of one or the other of these hypotheses. Our results with ovalbumin show that intrinsic molecular re-arrangement at this interface occurs only during the first five minutes, and that the progressive increase in film area thereafter is due to migration to, and adsorption at, the interface of protein which had escaped into the bulk phase. Some of the interesting biological consequences of this finding are discussed.

FUNDAMENTAL PARTICLES IN THE COSMIC RADIATION. J. G. RETALLACK. (Read March 8, 1954). Data from cosmic ray and big machine research of the last five years suggests that there are twenty-one fundamental particles with rest masses ranging from zero to greater than the mass of the proton. These particles are produced in very high energy nuclear collisions. It is suggested that these particles are not all funda-

mental, but that some are combinations of others. The current evidence will be presented and discussed.

SLIDE CULTURE OF TUBERCLE BACILLI. I. MEDIA AND METHODS. R. W. REED. (Read April 12, 1954). A modified slide culture technique is described for the cultivation of tubercle bacilli from untreated sputum specimens. Cultures are examined for growth under the low power after six and fourteen days incubation. Nearly two hundred cultures of sputum from sanatorium patients were positive either on slide culture or on Herrold's egg-yolk agar. Sixty-seven per cent were positive by both methods. However, 87% were positive by slide culture while only 80% were positive on Herrold's. The greater sensitivity of the slide method was confirmed by quantitative studies. Slide cultures were positive in an average of ten days, Herrold's agar cultures in thirty-one days. It is suggested that because of its technical simplicity and the rapid development of positive cultures, the slide culture method warrants further trial as a diagnostic procedure.

ORGANO-ANALYTICAL REAGENTS-AROMATIC HYDROXYLAMINES. G. D. LUTWICK and D. E. RYAN. (Read April 12, 1954). Aromatic hydroxylamines containing a carbonyl grouping α -to the oxime react much the same as cupferron but are stable to heat, light and air. These compounds are more sensitive to acids than cupferron; only titanium, zirconium, tin and vanadium are completely precipitated from five per cent acid solutions. Reaction involves inner complex formation except for tin; reduction of tin (IV) to tin (II) by the reagent seems a necessary prerequisite to precipitation.

THE GEL STRENGTH AND OPTICAL ACTIVITY OF CARRAGEENIN. D. A. I. GORING. (Read April 12, 1954). *Chondrus crispus* was extracted with 0.02 M aqueous sodium acetate at 100° C and the extract was separated into gel and sol fractions by addition of potassium chloride using Smith and Cook's method. Sub-fractionation of the gel fraction was then made in the same way. All fractions were converted to the sodium salt by dialysis. The sol fractions exhibited a marked decrease in both gel strength and specific rotation. The gel fractions showed a corresponding increase.

The whole plant was extracted with 0.02 M aqueous sodium acetate at 60° C, 100° C and 120° C successively and the gel strength and optical activity of the extracts were measured. The portions extractable at higher temperatures possessed lower gel strength and specific rotation. A control, obtained at 60° C, was also subjected to the higher temperatures used; this showed a relatively small change in specific rotation and gel strength.

It was concluded that at least two structurally different polysaccharides exist in *Chondrus crispus*, separable by means of potassium chloride and designated gel and sol components. Extracts made at 60° C were rich in the gel component. The sol component was concentrated in extracts made at higher temperatures.

A KINETIC AND THERMODYNAMIC STUDY OF INTRACELLULAR YEAST CATALASE. M. J. FRASER. (Read May 3, 1954). In a previous paper from this laboratory presented before this Institute, it was shown that the great increase in activity (and changes in various other properties) of intracellular yeast catalase that is brought about when yeast cells are

treated with various physical and chemical agents such as ultraviolet light, chloroform and propanol, is due to an "enzyme alteration". Enzyme alteration was defined as "a change in the properties of the enzyme itself resulting from the direct or indirect effect of the agents used, on the structure of the enzyme". In the present study the thermodynamic constants of the activation process, the experimental energy of activation (u), the heat of activation (ΔH^*), the entropy of activation (ΔS^*), the free-energy of activation (ΔF^*), for the catalase- H_2O_2 reaction and for the heat inactivation, for both unaltered and altered intracellular catalase, have been determined from kinetic data on the rates of these reactions measured at various temperatures. The ΔH^* and ΔS^* values for the unaltered catalase- H_2O_2 reaction are respectively 4,500 cal/mole and 11 cal/mole deg. Abs. *higher* than those for the altered catalase- H_2O_2 reaction, while the ΔF^* value is 1.7 Kcal/mole higher for the unaltered catalase- H_2O_2 reaction. The ΔH^* and ΔS^* values for the heat inactivation of the unaltered yeast catalase are respectively 20,600 cal/mole and 70 cal/mole deg. Abs. *lower* than those for the heat inactivation of the altered enzyme, while the ΔF^* is 23.0 Kcal/mole for both states of the catalase. These differences in the thermodynamic constants for the two states of intracellular yeast catalase show that the unaltered enzyme is structurally less specific, and such results are consistent with the hypothesis that catalase of the living yeast cell exists in a partially unfolded condition at some intracellular interface.

POSITRON ANNIHILATION IN METALS. R. E. GREEN and A. T. STEWART. (Read May 3, 1954). Positrons and electrons annihilate one another converting their total mass into energy. The energy usually appears in the form of two gamma rays which, if the pair is at rest, are emitted at 180° . If either of them is moving before annihilation, the angle observed between the gamma rays departs from 180° by an amount which depends upon the velocity of the moving particle. In a metal there are many freely moving electrons. When a positron enters such a substance, it rapidly loses nearly all of its kinetic energy by collisions, after which it annihilates with one of the electrons. From a measurement of the angular distribution of the emitted gamma rays, information about the average velocity of the annihilating electrons is obtained.

Apparatus to measure the angular distribution of gamma rays from positron annihilation has been set up and measurements have been made on a series of metals. Results of this experiment are compared with other experimental and theoretical methods of obtaining the velocities of free electrons in metals.

AGE DETERMINATION OF A SUBMERGED TREE STUMP. BY CARBON 14 ANALYSIS. A. L. CARTER. (Read May 8, 1954). Carbon in living matter is radioactive with a specific activity of about fifteen counts per min. per gram. This activity arises from the isotope carbon 14 which has a half-life of 5570 years. Thus the amount of carbon 14 diminishes with time after metabolism ceases.

An apparatus has been constructed to extract carbon from organic matter pure enough to measure this extremely small carbon 14 concentration. These measurements are taken with the carbon pasted on the cathode of a Libby screen-wall G.M. counter which is shielded with one inch of mercury, a ring of anticoincidence counters and eight inches of iron. The age of a tree stump situated twenty feet below the high tide level near Avonport, N. S. has been determined by comparing its carbon 14 concentration to that of modern wood.