BACTERIOLOGY

Chairman: Gordon Mallett, Eli Lilly

Gordon Mallett, Eli Lilly and Indiana University, was elected chairman for 1962

ABSTRACTS

Studies on the Increase *in vitro* of Mitotic Activity and Melangenesis in the RPMI HA # 5 (7113) Strain Melano. Judyth Vary and Sister M. Clare Francis, St. Francis College.—A 73rd generation un-pigmented melanoma, derived from a metastatic lesion in a human host and cultured in the Syrian hamster, was used in attempts to accelerate the proliferation of the melanoma *in vitro*, employing assays of the basic media #213 against controls of 213, Puck's, Shu, and ELH media. Several hundred variations of twelve amino acid concentrations, in correlation with fetal calf serum percentages of 2%, 5%, and 10% were tested. Although results are inconclusive at this date, indications suggest that specific concentrations of phenylalanine, alanine, and tryptophane influence from a slight to substantial extent the increase in mitotic activity of the melanoma. In some instances melanogenesis was increased to the point that some cells seemed to contain melanin in amounts noticeable under low microscopic powers. Tests with the *dopa* reaction revealed an increase in melanogenic activity in some cases.

The factors influencing accentuated mitosis and melonagenesis may provide a key in the control of this deadly cancer, since the absence or loss of such factors may reciprocally influence the proliferation and metastatic activity of this melanoma in an adverse manner. An area of future endeavor includes testing the influence of Ehrlich-derived ascites DNA; stock RNA; insulin; etc.

Cytochemical Changes Induced in Replicating Trachoma Virus by Metabolic Analogues. Morris Pollard, Lobund Laboratories, University of Notre Dame.—Replicating trachoma virus induces the same sequence of cytochemical changes in tissue cells as other members of the T-L-P family of viruses. When infected tissue cultures were periodically stained with acridine orange and observed with ultra violet light, the DNA virus particle in the cytoplasm was surrounded by an RNA matrix, from which mature DNA emerged.

Addition of antibiotics to the culture medium of infected cells interrupted the cytochemical sequence. At determined time intervals following infection, alterations in virus were induced as follows: aminopterin at time 0 interrupted replication at the RNA stage; 5 fluorouracil at time 14 hours induced formation of massed "abnormal" fraudulent RNA; and 5-fluoro-2' deoxyuridine at time 14 hours induced abnormal DNA-appearing material. An eclipse stage in replication of trachoma virus has been demonstrated by this intracellular chemical indicator system.

The Use of Peracetic Acid to Obtain Invertebrate Eggs for Gnotobiotic Studies. James P. Doll, P. C. Trexler, G. R. Bernard and Louise

LINDHOLM, Lobund Laboratories, University of Notre Dame.—Various techniques both chemical and mechanical have been used to procure bacteriologically sterile invertebrates, but not all methods have proved equally successful. Some do not render the egg completely germfree and antibiotics must be added which may interfere with certain studies; and in some instances the disinfecting agent will tend to dry out the egg which results in either death to the embryo or a maimed offspring.

Peracetic acid shows germicical activity against a broad spectrum of gram positive and gram negative organisms as well as fungi. It can be used and is effective in very low concentrations: as little as 0.001% in phosphate buffer and 0.020% in nutrient buffer will kill *E. coli and Micrococcus pyrogenes* within 10 minutes. It is also sporicidal: 1.0% of peracetic acid in the liquid phase will inactivate spores of *B. sterothermophilus* on glass beads within 30 seconds. Because of these properties and also because the residual products (water and oxygen) are non-toxic or can easily be washed out, the use of peracetic acid should find popularity among workers doing axenic studies.

In the present work, eggs of *Heterakis gallinae* (vector of *Histomonas meleagridis), Blatella germanica* (cockroach), *Drosophila melanogaster,* and *Artemia salina* were treated with various concentrations of peracetic acid and then tested for both bacterial sterility and for viability. The tests for microbial sterility were a modification and extension of the microbial control methods used at LOBUND Institute. The test for viability of *H. gallinae* was the ability to transmit blackhead to turkeys and grow in chickens; the ability of the embryo to hatch and maturate was the test of viability for the other invertebrates used. Results showed that in all instances the eggs could be effectively decontaminated and yet maintain viability of their embryos. Two of the germfree strains thus obtained were maintained for over a year and a half.

Dose-Response Relationships of X-Irradiated Germfree and Conventional Mice. BROTHER RAPHAEL WILSON, Lobund Laboratories, University of Notre Dame.—Dose-response curves for Lobund Swiss mice exposed to x-radiation were determined from data on 100 germfree and 166 conventional mice. The curves appear to be parallel sigmoid curves with that for the germfree mice about 50r to the right to that for the conventional mice. The data indicate that in the absence of microorganisms, the radiosensitivity of the mouse is reduced. In the mid-lethal range, 20-30% greater mortality occurs among conventional mice than among the germfree. Following exposure to fatal dosage of radiation, the germfree mouse lives longer than its conventional counterpart. Plotting survival time against dosage for conventional mice yields a curve that drops sharply range a survival time of 14 days to a plateau of about 4 days in a dose range of 500-2000r. The corresponding survival time curve for germfree mice begins with a short plateau at about 12 days and drops gradually to about 6 days. The use of germfree animals enables the study of radiation syndrome in the absence of post-irradiation infection.