

ABSTRACTS OF PAPERS PRESENTED AT THE 1961 MEETINGS OF THE KENTUCKY-
TENNESSEE SOCIETY FOR MICROBIOLOGY, OCTOBER 27 AND 28, 1961, DEPARTMENT OF
MICROBIOLOGY, UNIVERSITY OF KENTUCKY, LEXINGTON.

President, Ralph F. Wiseman,
University of Kentucky
Secretary, Raymond W. Beck,
University of Tennessee

- 1. Electron microscopy of internal structure of aerobic actinomycetes.** O. F. Edwards, H. H. Haines and M. Hochkins, Department of Microbiology, University of Kentucky, Lexington.
Unsectioned preparations, examined with the electron microscope, show only the peripheral features of the actinomycetes. After the fragments of the cultures are fixed with Osmium tetroxide, embedded in plastic and sliced with the Porter-Blum microtome, it is possible to demonstrate cell walls, septa in the filaments and internal structures that some workers interpret as chromatinic material. It is certain that these branched microorganisms are multicellular rather than unicellular. The branches arise by protrusions from a cell and the protoplasm flows, unobstructed by a septum, into the developing filament.
- 2. Problems presented by the preparation of thin sections when applied to bacteria.** H. Haines, O. F. Edwards and M. Hochkins, Department of Microbiology, University of Kentucky, Lexington.
A discussion of methods for the fixation, dehydration and embedding of bacterial specimens to be used with the Porter-Blum microtome.
- 3. Characterization of a new species of Bacteroides, with special reference to its fermentation end-products.** Arthur P. Harrison, Jr., Division of Molecular Biology, Department of Biology, Vanderbilt University, Nashville.
A large gram-negative, obligately anaerobic rod of avian intestinal origin has been studied extensively. Its most striking features are large size (1.5 to 2 micra x 5 to 20 micra) and strong fermentative ability (17 out of 22 carbohydrates and polyhydric alcohols are fermented). One of the 4 strains shows the bizarre morphological forms often associated with this genus. Great sensitivity is manifest *in vitro* toward penicillin as well as tetracycline, aureomycin, and chloramphenicol. The organism has exacting nutritional requirements and has not been cultivated on synthetic media, but like other *Bacteroides* is easy to cultivate on commercially available dehydrated media such as yeast extract. The pentoses and disaccharides are preferred to the usual monosaccharides and are preferred to the usual monosaccharides and polyhydric alcohols. The fermentation spectrum is altered by hypophosphatization, and this alteration is not uniform but varies from strain to strain. Apparently, great genetic diversity occurs in this species. Approximately 75% of the fermented glucose may be accounted for as acetic acid (10%), propionic acid (20%) and dextralactic acid (45%). The remainder of the fermented glucose has not been accounted for, but negligible quantities of levo-lactic acid, butyric acid, ethanol, acetone, and gas are produced.
- 4. Alterations in the hemoglobin pattern of chicks infected with *Salmonella pullorum*.** Ronald A. LeClair and D. Frank Holtman, Department of Bacteriology University of Tennessee, Knoxville.
Recently hatched White Leghorn cockerels, when injected with a lethal dose of viable *Salmonella pullorum* cells exhibit reduced hemoglobin levels. The infection is accompanied by anorexia and a reduction in the amount of glycine in the blood serum which suggests its failure to be absorbed from the gastrointestinal tract. The administration of glycine by intraperitoneal injection or in diets fortified with high levels of glycine prevents reduction in hemoglobin levels. Similar results can be obtained by feeding chick basal ration fortified with pyridoxal phosphate, but not by a glycine-deficient diet containing pyridoxal phosphate.
- 5. Observations on an incomplete lecithinase produced by *Pseudomonas* species.** Janice L. Bates and Pinghui V. Liu, Department of Microbiology, University of Louisville School of Medicine, Louisville, Kentucky.
Pseudomonas aureofaciens strain B-2265 and *Pseudomonas fluorescens* strain 22/3 when streaked on an egg yolk agar plate as separate colonies did not produce a lecithinase reaction around their respective colonies. However, when streaked close together a lecithinase reaction occurred in an area midway between the two colonies. The two factors involved in the reaction were obtained in crude cell free filtrates using the cellophane plate technique. When tested in a 5% egg yolk solution neither the B-2265 filtrate nor the 22/3 filtrate produced a reaction but a combination of the filtrates produced a titer of 1:64. Liberation of choline was detected both quantitatively and qualitatively. The B-2265 factor is non-dialyzable and antigenic while the 22/3 factor is dialyzable and non-antigenic. Antisera produced against the B-2265 filtrate neutralized not only the lecithinase reaction produced by the combined factors but the lecithinase reactions of other closely related species such as *P. aureofaciens*, *P. chlororaphis* and *P. fluorescens*. They did not neutralize the lecithinases of *P. aeruginosa*, *P. pseudomallei* or *P. reptilivora*. Likewise antilecithinase sera of *P. aureofaciens*, *P. chlororaphis* and *P. fluorescens* neutralized the lecithinase reaction of the combined filtrates of B-2265 and 22/3 but antilecithinase sera of *P. aeruginosa*, *P. pseudomallei* and *P. reptilivora* did not. This observation of incomplete lecithinase action underscores the fallibility of determining species within this genus solely on the ability of the organism to produce or not produce an enzymatic or biochemical reaction.
- 6. Transfer of UV (2537 A) resistance in mating strains of *Escherichia coli*.** James C. Copeland and Howard I. Adler, Biology Division, Oak Ridge National Laboratory, Oak Ridge.
During mating of a VHF (very high frequency) strain of *E. coli*, radiation resistance to UV may be passed from a resistant donor to a sensitive recipient producing progeny with varying radiation resistance. From such a cross, three classes of progeny are produced. One class is like the resistant donor, and the third class is intermediate between the donor and the recipient. The data indicate that the intermediate class may be further subdivided. The occurrence of these three classes of progeny is dependent upon the regions of donor genetic material incorporated. The data is consistent with the idea that more than one gene is responsible for the difference in radiation sensitivity between the parental strains. The relation of other biological parameters to

- the radiation response of the parental types and the progeny were discussed.
- 7. Genetic analysis of X-ray resistance in *Escherichia coli*.** Howard I. Adler and James C. Copeland, Biology Division, Oak Ridge National Laboratory, Oak Ridge.
Matings between an X-ray resistant VHF (very high frequency) donor strain of *E. coli* and a sensitive recipient were made. Many of the progeny are as sensitive as the original recipient parent, a few are as resistant as the donor parent, and a large group are of intermediate resistance. An analysis of the data suggests that more than one distinct class may be present in the intermediate group. The difference in radiation sensitivity of the parent strains may be ascribed to a group of genes, some of which seem to be located on the bacterial chromosome between the genes controlling histidine and proline synthesis. Comparison of results from X-ray and ultraviolet inactivation studies suggest that many of the same genes are involved in determining response to both kinds of irradiation.
 - 8. An extracellular proteolytic enzyme of *Streptococcus faecalis* var. *liquefaciens*.** L. Howard Moss III, J. Orvin Mundt and Raymond W. Beck, Department of Bacteriology, University of Tennessee, Knoxville.
Streptococcus faecalis var. *liquefaciens* produces a rennin curd in milk which subsequently is digested. A sample fractionation of the crude enzyme preparation yielded a single active fraction, which was thermally inactivated between 60 and 65 C, and which showed maximum proteolytic activity between pH 7.0 and 7.5. The Michaelis-Menten constant on a casein substrate was 0.1 per cent. L-tyrosyl glycine was hydrolyzed at pH 7.2. Proteolytic activity was increased markedly by addition of free arginine to milk cultures of the organism.
 - 9. The effect of a nitroimidazole on the microbiology of the vagina.** Emil Kotcher, Carolyn A. Frick, and Laman A. Gray, Departments of Microbiology and Obstetrics and Gynecology, School of Medicine, University of Louisville, Louisville.
A systemic drug, 1-beta hydroxyethyl, 2-methyl, 5-nitroimidazole, "Flagyl", was given to women with vaginal trichomoniasis because of its demonstrated activity against *Trichomonas vaginalis*. On oral doses of one gram per day for 10 days, a series of patients showed 1) a decrease in the incidence of *T. vaginalis* from 100% to 7%, 2) a slight increase in the number of patients with *Candida albicans*, 3) an increased incidence of patients with Doederlein's bacillus from 61.4% to 79.4%, 4) an increased incidence of patients with a low vaginal pH, and 5) a lowered incidence of clinical vaginitis.
 - 10. Cervical cell inclusion bodies and viral infections of the cervix.** Emil Kotcher, Laman A. Gray, Quinton C. James, Carolyn A. Frick, and Doris W. Bortoff, Departments of Microbiology and of Obstetrics and Gynecology, School of Medicine, University of Louisville, Louisville.
Almost 8,000 Giemsa-stained cervical smears were examined for basophilic inclusion bodies and eosinophilic elementary bodies. The smears were obtained from women in the reproductive and post-menopausal years. Positive smears were found in 218 women, 2.8%. The blue-staining inclusion bodies varied in size up to 10 micra. The red-staining elementary bodies are much smaller, about 0.2 micra, and may be found extracellularly as well as intracellularly. Cervical smears from positive women, when stained with D'Antoni's iodine stain, showed dark-brown intracytoplasmic bodies indicative of glycogen. The inclusion and elementary bodies are deemed to be the virus of inclusion conjunctivitis. One strain of this viral agent has been isolated in the yolk sac of embryonated chicken eggs. Giemsa-stained yolk sac smears showed eosinophilic elementary bodies in clusters intracellularly, and individually, extracellularly. These elementary bodies are similar to those found in the epithelial cells and leucocytes of cervical smears.
 - 11. Cultural and serological studies on *Haemophilus vaginalis*.** Donna L. Redmond, and Emil Kotcher, Department of Microbiology, School of Medicine, University of Louisville, Louisville.
Fluorescein-tagged antisera were prepared for two strains of *Haemophilus vaginalis* and one strain of *H. agyptius*. The specificity of the antisera was determined by the direct technique with homologous and heterologous strains of *H. vaginalis*, homologous and heterologous strains of *H. agyptius* and strains of *H. influenzae*, types A, B, C, D, and E. Cross-reactions did not occur with the *H. vaginalis* antisera and the strains of *H. influenzae* and *H. agyptius*, nor with vaginal diphtheroids, lactobacilli, *E. coli*, *Klebsiella-Aerobacter* group, *Staph. aureus*, *Streptococcus viridans*, and *X. gonorrhoeae*. Cross-reactions did not occur with *H. agyptius* antiserum and a strain of *H. influenzae* but not with strains of *H. vaginalis*. To test the clinical application of the *H. vaginalis* fluorescent antiserum, 26 patients with varying degrees of vaginitis were studied by 1) wet mounts for clue cells, 2) culture on Canning's medium, 3) vaginal smears stained with the *H. vaginalis* anti-serum, and 4) vaginal smears stained by Gram's method. A high positive correlation was found between the cultural and immunofluorescent technique.
 - 12. The specificity of the inhibition of migration of leukocytes of tuberculous subjects by products of microbial growth.** E. H. Gerlach and M. Scheraga, Department of Microbiology, University of Kentucky, Lexington.
The hypersensitivity of leukocytes of normal and of tuberculous subjects was determined by a migration inhibition technique. The specificity of the reaction was determined using filtrates of *M. tuberculosis*, *M. bovis*, *M. phlei*, *E. coli*, and *Histoplasma capsulatum*. Ten normal subjects exhibited no specific leukocytic sensitivity to any of the filtrates. Twelve tuberculous subjects showed doubtful or nonspecific sensitivity to tuberculin. One of these subjects exhibited a specific leukocytic sensitivity to an *E. coli* filtrate and one to histoplasmin. Of the thirty-nine remaining subjects with specific leukocytic sensitivity to tuberculin only one subject showed a specific leukocytic sensitivity to an *E. coli* filtrate. Three others exhibited a specific leukocytic sensitivity to histoplasmin.
 - 13. Uptake, concentration, and release of cobalt and cesium by soil fungi.** Martin Witkamp, Health Physics Division, Oak Ridge National Laboratory, Oak Ridge.
Aspects of the influence of microorganisms on distribution and turnover of minerals was studied by growing soil fungi on media containing radioactive cobalt or cesium. Uptake of Co^{60} and Ce^{137} by *Trichoderma viride* in liquid media was maximal after 7 and 16 days respectively and directly proportional to the isotope concentration. Low and high pH of the media stimulated cobalt uptake, cesium uptake was slightly higher at high pH. Concentration factors for various soil fungi in liquid media ranged from 9 to 63 for cobalt and 4 to 27 for cesium. On agar media concentration factors in aerial mycelium of *Trichoderma* and *Mucor* sp. were 1.7 for cobalt, and 15.7 and 5.4 for cesium indicating better translocation of cesium than of cobalt especially in *Trichoderma*. Release of cobalt and cesium after one day leaching with water averaged from fresh, frozen, and dried mycelium 9 and 19, 89 and 97, and 85 and 96% respectively. In the field this release was masked by absorption of 2% of the released isotopes by a 1.5 cm layer of leaves. Microorganisms retain 30% of the cobalt and 38% of the cesium in leaves that otherwise would be removed by leaching.
 - 14. Cultivation of vaccinia virus in sheep kidney cell cultures.** P. S. Sarma (P. Subramanyam), S. Divakaran and P. Vinodraj, Department of Microbiology, University of Kentucky and Pasteur Institute of Southern India, Coonoor, India.
Attempts were made to find a suitable tissue for the preparation of cell monolayers for the cultivation of vaccinia virus and for the titration of the virus and its antibodies. Kidneys procured from freshly slaughtered healthy young sheep were found suitable for this purpose. The cultures were easy and economical to prepare and

supported the multiplication of the virus well. They could be used for the titration of the virus and its antibodies. The sensitivity of sheep kidney cell cultures to virus was comparable to that of chorioallantoic membranes of chicken embryos. Preliminary trials indicated that the sheep kidney cell culture virus could be freeze-dried. Preliminary studies on the antigenicity of the virus propagated in sheep kidney cell cultures indicate a potential use of these cultures for the preparation of a vaccine against smallpox.

15. *Growth of penicillin-resistant and penicillin-sensitive strains of Staphylococcus aureus.* Wendall Allen and Ilda McVeigh, Biology Department, Division of Bacteriology, Botany, and Zoology, Vanderbilt University, Nashville. Ten strains of naturally penicillin-resistant *Staphylococcus aureus* (obtained from patients), two *in vitro* derived resistant strains, and two sensitive strains were grown at 37°C in Antibiotic Assay broth, and viable cell determinations were made at intervals. From those data, growth curves were plotted for each of the strains. The curves for the naturally penicillin-resistant and the sensitive strains are very similar. Little, if any, lag in growth of these strains occurred on transfer from maximum stationary-phase cultures to fresh medium. They grew at approximately the same rate during the logarithmic growth phase, which lasted for 3 to 4 hours, and, during the maximum stationary phase, about the same number of cells was present per ml in cultures of each of these strains. In contrast, the *in vitro* derived resistant strains underwent a lag of 2 to 6 hours on transfer to fresh medium and grew at a slower rate during the logarithmic growth phase. However, during the maximum stationary phase, which occurred after an incubation period of 24 to 32 hours, the cell titers were approximately the same as those of the naturally resistant and the sensitive strains. When grown in competition with either of the sensitive strains in Antibiotic Assay broth in the absence of penicillin, one of the naturally resistant strains persisted for 14 successive subcultures without any apparent change in ability to tolerate the antibiotic.
16. *Penicillinase activity in rumen paracolon bacteria.* Phyllis I. Warren and Ralph F. Wiseman, Department of Microbiology, University of Kentucky, Lexington. Recent reports indicate that rumen paracolon bacteria may be responsible for the inactivation, after about one week of treatment, of penicillin used in controlling bovine bloat. This study was designed to determine whether rumen paracolons from treated and untreated animals are capable of producing penicillinase, *in vitro*. Thirty-three paracolon isolates from the rumen content of a steer which had never received penicillin, and forty isolates from a steer which was receiving 150 mg of penicillin daily, were grouped according to the International Bulletin of Bacteriological Nomenclature. Three isolates from each steer, representing the three predominating "species", were assayed for penicillinase activity. Penicillinase was not detected in the cell free filtrates of the isolates, even though the isolates were previously grown in a medium containing penicillin. Penicillinase activity was demonstrated in dried cell material, but this activity was dependent upon prior exposure, *in vivo* or *in vitro*, to penicillin.
17. *Identity of bacteria producing colonies on coliform plating media.* Virginia L. Colley and J. Orvin Mundt, Departments of Bacteriology and Food Technology, University of Tennessee, Knoxville. Coliform bacteria (species of *Escherichia* and *Aerobacter*) produce atypical as well as typical colonies on violet red bile and desoxycholate agars. Members of the genus *Pseudomonas* and unidentified, lactose fermenting rods constitute a large minority of both the typical and atypical colonies selected for identification.
18. *Optimum conditions for production of colominic acid.* G. T. Barry and J. D. Hamm, University of Tennessee Memorial Research Center, Knoxville. Colominic acid, a polymer of N-acetylneuraminic acid,

is elaborated by strains of *E. coli* which possess a K1 serotype. (Barry, et al., *Nature*, 1960, 185, 597.) As the amount of colominic acid recovered from culture supernates is low, a study was made to determine optimum growth conditions for production of this material. The effect which pH, temperature, nutrients and time of harvesting has on the yield were evaluated. Optimum yields of colominic acid are obtained when the bacteria are grown in media consisting of 1.5-3.0% glucose, 1.0-2.0% casamino acids (Difco) and 0.01 M phosphate buffer maintained at pH 7.5 at 37°C with aeration. At the end of the growth period (18 hours) the culture is aerated for an additional 30 hours. The yield of 20 mg. of colominic acid per 100 ml. of culture obtained by this procedure is double that previously reported (Barry, J. Exp. Med., 1958, 107, 507). Addition of yeast extract or increased phosphate does not enhance the yield. Thus, an improved method for the production of colominic acid was devised which employs a simplified growth medium. (Supported by a grant from the United States Public Health Service.)

19. *Type-specific antibodies against streptococcal M protein.* Nye P. Lowry and Robert W. Quinn, The Department of Preventive Medicine and Public Health, School of Medicine, Vanderbilt University, Nashville. The technique ordinarily used to demonstrate the presence of antibodies against the type-specific, M protein of the hemolytic streptococcus has been the bactericidal test. This test is difficult to perform and does not lend itself well to the testing of large numbers of sera. The present study describes a new precipitin technique for the detection of anti M antibodies. Chicken erythrocytes are coated with type-specific, M protein. In the presence of type-specific, anti M antibody the erythrocytes agglutinate. The results to date indicate that this antibody appears after infection due to group A streptococci. The type-specific antibody can be absorbed out of a patient's serum by homologous type-specific M protein. Type-specific antibodies against streptococcal types common in the Nashville area have been demonstrated in the serum of from 20-25% of all subjects tested.
20. *Studies of so-called non-typable group A streptococci.* J. T. Jones and Robert W. Quinn, Department of Preventive Medicine and Public Health, Vanderbilt University, School of Medicine, Nashville. Several group A non-typable strains of streptococci were recovered from school children in Nashville during the school year 1960-61. The purpose of this experiment was to determine whether or not these so-called non-typable streptococci actually contained M protein. The M protein is supposedly absent if the strain is truly non-typable since the M antigen gives each strain its type specificity. Other factors which might have influenced the production of M protein were the possibility that M protein might be present in such small quantities that it would be insufficient to give a positive test, and the possibility that these strains produced proteinase which destroyed the M protein. The results showed that one non-typable strain was capable of stimulating anti M antibody production in the rabbit, the other was not. Both produced proteinase. Serial mouse passage increased the virulence of the strains but M-protein production was not influenced. This experiment indicates some "non-typable" strains of group A streptococci do have M protein and are capable of stimulating antibody production in the rabbit and also in the patient.
21. *The hematopoietic response of the host to the injection of cortisone followed by endotoxin challenge.* Randall T. Jones and D. Frank Holtman, Department of Bacteriology, The University of Tennessee, Knoxville. It has been observed that the intraperitoneal injection of both purified and crude endotoxin preparations was followed by a marked reduction in peripheral leukocytes at six and twelve hours postinjection. Treatment with cortisone acetate one hour before endotoxin challenge was found to partially correct the leukopenic response of the intoxicated host. It is suggested that the effect exerted by cortisone upon the hematopoietic system of

the host may be a significant factor in the protective role of the steroid against the lethal action of endotoxin.

22. *The effect of PPD on the *in vitro* migration of leukocytes from guinea pigs passively sensitized to OT.* Roger W. Johnson and M. Scherago, Department of Microbiology, University of Kentucky, Lexington. Delayed skin hypersensitivity to tuberculin was passively transferred from guinea pigs experimentally infected with *Mycobacterium tuberculosis* H37Rv to normal guinea pigs by means of peritoneal exudate cells. The *in vitro* migration of leukocytes from the passively skin sensitized guinea pigs was not significantly inhibited by a concentration of PPD shown to inhibit the migration of the leukocytes did not vary significantly from that of leukocytes from normal guinea pigs.
23. *The pigments of Microsporium cookei.* Guy W. Koehne, Frederick T. Wolf and Ernest A. Jones, Department of Biology, Division of Molecular Biology, and Department of Physics, Vanderbilt University, Nashville. It has been suggested by previous workers that the pigments of dermatophytes are polyhydroxyanthraquinones, but the evidence for this is meager. The dermatophyte, *Microsporium cookei*, produces a purple pigment on Sabouraud's agar. By extraction with acetic acid, partitioning into chloroform, and chromatographing the concentrated extract on paper, using chloroform: petroleum ether 7:3, it was found that the pigment mixture includes a purple component and a bright yellow component. Both pigments are identical with metabolic products of *Trichophyton rubrum*. Both pigments are pH indicators and redox indicators. Quantities of the pigments from *M. cookei* were separated by column chromatography on sucrose, eluted, concentrated and prepared in solid form. From evidence derived from infrared absorption spectra, it is concluded that both pigments are polyhydroxymethyl anthraquinones. Both pigments must have at least one beta substituent on each of the lateral rings, and must have at least one alpha hydroxyl and one beta hydroxyl. The yellow pigment is fluorescent, and must have two alpha hydroxyls in the 1, 4 positions.
24. *A deoxyribonucleic acid separation procedure for use with mouse neoplasms.* William W. German and Anna D. Dulaney, Division of Pathology and Microbiology, University of Tennessee College of Medicine and City of Memphis Hospitals, Memphis. Investigation of the biological and chemical activity of deoxyribonucleic acid (DNA) is presently of active scientific interest. Such studies should require preparations of nucleic acids which may be standardized and evaluated chemically with minimal alteration of their biological activity. Infectious DNA has recently been reported for SE polyoma virus by DiMayorca et al. who utilized essentially the phenolic procedure described by Kirby. Evidence for such activity was based on the fact that untreated preparations were infectious while DNAase removed all infectivity. These procedures have been

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stressed both in lectures and in the daily workshop.

The Institute registration fee is \$10.00. The tuition fee is \$100.00 for each of the three sessions. Partial tuition scholarships are available to academic personnel. It is hoped that, as last year, arrangements for 30 or 40 awards covering tuition, maintenance and travel for academic personnel will be possible through National Science Foundation aid.

All Institute facilities, including dormitory accommodations are air-conditioned. The tentative fee for university housing and meals is \$50.00 for each session.

modified by us for separation of DNA from mouse sarcomas. Basically, homogenized tissue was first treated with sodium p-amino salicylate followed by 90 per cent phenol. The DNA present in the aqueous phase was precipitated once with alcohol and redissolved in phosphate buffer, pH 7.2, followed by six ether extractions. The final steps were dialysis followed by lyophilization. This material contained a high DNA content and as low as 0.4 per cent protein. A chemical evaluation of the authors' procedure, as compared to that of Kirby and its modification by DiMayorca et al. will be presented. (Supported in part by a grant from the National Cancer Institute, National Institutes of Health, United States Public Health Service.)

25. *Pseudomonas infection in the mouse colony at Oak Ridge National Laboratory.* John M. Woodward, Biology Division Oak Ridge National Laboratory and the University of Tennessee, Knoxville. Death of mice within 4 to 7 days following X-irradiation was correlated with a high incidence of *Pseudomonas* carriers in the mouse population. It was shown that dissemination of this organism occurred primarily via the drinking water as a result of unsanitary watering procedures. The transfer of *Pseudomonas* from mouse to water to mouse appeared to enhance the spread of this contaminant. Improvement in sanitation during the bottle washing and filling operations, coupled with the addition of 10 P.P.M. available chlorine to the drinking water, reduced the distribution of *Pseudomonas* from this source. Other controls included the killing of *Pseudomonas* carriers in the colony and changes in the operating procedures which improved sanitary conditions in all areas of the animal facilities. These measures have reduced drastically the incidence of *Pseudomonas* in the mice although the organism has not as yet been eliminated from the colony.
26. *The control of Pseudomonas in a mouse breeding colony.* Raymond W. Beck, Cumberland View Farms, Clinton, Tennessee, and the University of Tennessee, Knoxville. Reports that early death in irradiated mice was attributed to infection with *Pseudomonas* led to an investigation of the incidence and means of control of the organism in a mouse breeding colony which supplies inbred mice to laboratories engaged in radiobiological research. *Pseudomonas* was found throughout the colony including a section operated as specific pathogen free (SPF colony). Efforts to rid the SPF colony of the organism by killing *Pseudomonas* carriers or by dosing the drinking water with Polymyxin B were unsuccessful. The bottle washing and filling operation was implicated as a means of spread of *Pseudomonas*. The incidence of the organism has been drastically reduced by the routine use of disinfectant in the bottle wash area, by improving the bottle filling and stoppering procedure to avoid contamination of clean bottles, by the addition of 10 P.P.M. available chlorine to the drinking water, and by changing the water bottles on a two or three times per week schedule.

Institute Session enrollments are limited to 60 persons each in the Gas Chromatography and the First Infrared Session (14-18 August), and to 80 persons during the Second Infrared Session (20-24 August). Participants may enroll for either of the 14-18 August Sessions, and/or for the 20-24 August Session. The Institute registration list is usually rather equally divided between scientific personnel from industrial laboratories, governmental research laboratories and academic institutions.

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