**986** THE PATHOGENESIS OF COMBINED MURINE CYTOMEGALOVIRUS AND ESCHERICHIA COLI K1 INFECTION IN MICE. J.F.Bale, Jr., E.R. Kern, J.C. Overall, Jr. and L.A. Glasgow. University of Utah College of Medicine, Department of Pediatrics, Salt Lake City, Utah.

Cytomegalovirus (CMV) infections have been associated with increased susceptibility to serious bacterial infections in transplant patients and in young infants. To explore further the mechanisms by which CMV infection alters susceptibility to bacterial infections 3-week-old Swiss-Webster mice were inoculated intraperitoneally with murine CMV (MCMV) and then were challenged intranasally with E. coli K1. In MCMV-infected animals, challenge with E. coli resulted in enhanced mortality (70-90% vs 0-20% in controls). Mortality was greatest when animals were infected with E. coli on days 1 or 3 after MCMV inoculation. Clearance of E. coli from blood and organs was impaired on day 3 of the MCMV infection. Forty-eight hours after E. coli inoculation, MCMV-infected animals had greater bacterial counts than control animals in blood ($10^{4.5}$ E. coli/ml in MCMV-infected vs $10^{1}$ controls), in lung ($10^{7}$ E. coli/g vs $10^{3.5}$), in spleen ($10^{4.5}$vs $10^{1}$) and in liver ($10^{4.5}$ vs $10^{1}$). In addition, MCMV-infected animals exhibited a diminished inflammatory response to a subcutaneously implanted sponge containing $10^{8}$ E. coli (mean of 740 WBC/$mm^3$ in MCMV-infected vs 5783 WBC/$mm^3$ in controls). These results indicate that infection with MCMV enhances susceptibility to intranasal E. coli infection and suggest that acute CMV infection alters host inflammatory response to a bacterial stimulus.

**987** COMPARISON OF OUTER MEMBRANE PROTEINS OF TYPE B AND NONTYPE B HAEMOPHILUS INFLUENZAE Stephen J. Barenkamp, Robert S. Munson, Jr., Virgil M. Howie and Dan M. Granoff, Washington Univ. School of Medicine, St. Louis, MO

We have proposed a subclassification scheme for H. influenzae type b (Hib) based upon distinctive and reproducible strain differences in the SDS-polyacrylamide gel electrophoresis patterns of outer membrane (OM) proteins. Of 49 invasive isolates from patients hospitalized in St. Louis (StL), 92% could be assigned to 1 of 5 subtypes (1L,1H,2L,2H,3L). In the present study, we used this system to subtype 86 invasive isolates from patients hospitalized in 12 other states. The results were compared to the subtypes in StL, and to the OM protein patterns of nontype b Haemophilus isolates from middle ear (14 isolates), or blood/CSF (4 isolates). The overall distribution of Hib subtypes in other areas of the U.S. was similar to St.L ($p>.35$). However, there were possible regional differences: Isolates from New Orleans had a higher proportion of 3L strains than the rest of the U.S.,6/15 (40%) compared to 16/120 (13%), $p<.01$. Also, 3 of 11 apparently unrelated type b isolates from Denver had identical OM protein patterns not identified in 124 other isolates ($p<10^{-8}$). Nontype b isolates (middle ear or invasive) had OM protein patterns different from the common type b patterns, and exhibited much greater variability. Thus, type b isolates causing invasive infections in the U.S. appear to be derived from a small number of distinctive clones. Nontype b isolates are genetically distinct from type b strains, and are more heterogenous. These findings may have important epidemiologic and immunologic implications.

**988** AMPICILLIN (A)/NAFCILLIN (N) SYNERGY AGAINST AMPICILLIN-RESISTANT HAEMOPHILUS INFLUENZAE TYPE B ($Hib^R$). William J. Barson, Robert J. Fass, Frank A. Kapral, Robert L. Brawley, and Milo D. Hilty, The Ohio State University College of Medicine Depts. of Peds., Med., and Med. Micro. and Children's Hospital Dept. of Peds., Columbus.

Three infants with $Hib^R$ bacteremic infections had good clinical responses to A and N therapy. This observation prompted in vitro evaluation of this combination against $Hib^R$ clinical isolates. Minimal inhibitory concentrations (MICs) in µg/ml of A and N alone and in combination were determined by a microdilution method for 5 $Hib^R$ isolates; A: 8-32, N: 8-16, A/N: 0.5-1/1-2 at a $10^4$ colony forming unit/ml inoculum. Results indicate synergy since the MIC of each drug alone is at least 4 times greater than the MIC of the drug in combination. The proposed mechanism of A/N synergy is N inhibition of β-lactamase which protects A from hydrolysis allowing it to exert its antibacterial effect. A spectrophotometric assay of β-lactamase activity was used to study this proposal. The supernatant of sonified $Hib^R$ cell preparations was the β-lactamase source; A served as substrate; and N, methicillin(M) and oxacillin(O) were studied as potential inhibitors. The hydrolysis of A in the absence and presence of the inhibitors obeyed Michaelis-Menten kinetics. N, M, and O acted as purely competitive inhibitors of A hydrolysis with N being the most effective and O the least as evidenced by $K_I$ values of $7.10 \times 10^{-2}$, $3.16 \times 10^{-1}$, and 3.35 µM respectively. These findings provide an explanation for the observed in vitro A/N synergy which in vivo may have been responsible for favorable outcomes in 3 infants with $Hib^R$ bacteremic infections treated with A and N.

**989** INTRINSIC BACTERICIDAL CAPACITY OF NEONATAL MONONUCLEAR PHAGOCYTES (MP) AND POLYMORPHONUCLEAR CELLS (PMN) AGAINST GROUP B STREPTOCOCCI (GBS) TYPES Ia AND III. Ingeborg D. Becker, Octavio M. Robinson, María L. Collado and Roberto R. Kretschmer. Division of Immunology. Subjefatura de Investigacion Cientifica, CMN-IMSS and Instituto de Investigaciones Biomédicas, UNAM. Mexico City, México.

The intrinsic bactericidal capacity of MP and PMN of 16 normal newborn infants and 10 healthy adults against GBS-Ia and III prototype strains was measured in vitro using optimal opsonic (single batch fresh pooled human serum) and leukocyte:bacterial (1:10) ratios. Interstrain variability of intrinsic bactericidal capacity was further evaluated exposing MP and PMN of three normal newborns simultaneously to five different GBS-III strains. Of all cell:bacterial combinations, only newborn PMN were found to have a significantly ($p<0.05$) diminished bactericidal index (B.I.) (64.6% ± 8.1%) (mean ± s.e.) vs GBS-III when compared to adult PMN (87.2% ± 4.3%). However, 25% of all newborn PMN were significantly less bactericidal (B.I. ≤ 40%) towards both GBS-Ia and GBS-III by the Moses test for extreme reactions ($p<0.05$). Of the three sets of newborn leukocytes confronted with five strains of GBS-III, PMN of one revealed diminished B.I. against two strains, while the other two sets were bactericidally normal against all five strains. In addition to critical deficiencies in opsonic factors, newborns may also have intrinsic cellular bactericidal deficiencies against some GBS strains and this may constitute another risk factor for contracting GBS disease.

**990** THE RELATIONSHIP OF AGE OF ONSET OF OTITIS, ALLERGIC HISTORY, AND PREMATURITY TO RECURRENT OTITIS MEDIA. Stephen Berman, Jeanine King, Jan South. Spon. by William Hathaway. University of Colorado Health Sciences Center, School of Medicine, Department of Pediatrics, Denver, Colorado.

This retrospective study of OME was undertaken to examine the relationship between age of onset of otitis, allergic history, prematurity, and recurrent and/or chronic otitis media. Included in the study were 341 children seen at the ENT Clinic with a history of acute serous, secretory, or chronic otitis media, born after January 1, 1974. A total of 1786 episodes were documented in charts. Otitis media with serous effusion (SOM) accounted for 36% of the episodes and otitis media with purulent effusion (AOM) accounted for 60%. Four percent of the episodes had SOM in one ear and AOM in the other. AOM developed in 34% of the episodes of SOM. Mean duration of SOM and AOM from onset to total resolution was 43 and 18.2 days respectively. In those episodes of SOM that developed acute otitis, the mean duration prior to becoming an acute otitis was 28.3 days. The durations of the hearing loss associated with SOM and AOM were 56.1 and 23 days respectively. Positive history of allergy increases the risk of recurrent otitis media in the second year of life. Prematurity does not appear to increase the frequency of otitis media in the first two years of life. Frequency of otitis media following the onset of OME is not affected by the timing of age of onset within the first two years. Timing of exposure to upper respiratory tract season appears to be a significant variable in determining age of onset and frequency of OME.

**991** THE USE OF PLASMA INFUSION FOR NEONATAL SEPSIS Robert Bortolussi, Andrew C. Issekutz, Dorothy R. Barnard (Spon. by Richard B. Goldbloom). Dalhousie University, Izaak Walton Killam Hospital for Children, Halifax, Canada.

To assess the value of fresh frozen plasma (FFP) infusion to neonates with sepsis we obtained blood samples from 20 infants with suspected sepsis before and after i.v. FFP infusion (20ml/kg). Classical (CCP) and alternative (ACP) complement hemolytic activities, opsonic activity (OA) against the infecting organism and coagulation factors VIII and XIII activity and antigen were evaluated. Sera from mothers of septic infants, from matched control infants, and from recalcified FFP were compared to adult pooled serum. Of the 20 study infants, 7 had sterile blood and CSF cultures while 13 had isolates from blood, CSF or middle ear, including 7 group B streptococcus and 2 L. monocytogenes. Low CCP and/or ACP activity, present in half of the patients was corrected in all after plasma infusion ($p<.001$). ACP and CCP activities were normal in FFP. Depressed OA to the infecting organism ($p<0.01$ vs control infants) was seen before FFP. After FFP, OA improved (7/12) or was unchanged in all but 1 culture +ve infant. Compared to adult serum, OA was similar in control infants (median 109%) and in FFP (104%) but lower in mothers of infected infants (71%). Increased ratios of Factor VIII and XIII antigen/activity compatible with proteolysis and/or mild DIC were present prior to FFP and approached normal 20 hours later. We conclude that FFP infusion to septic infants is safe. The observed changes in complement and opsonic activity may benefit the depressed humoral host defense system. (MRC grant No. 210)