1039 IN VITRO LYMPHOCYTE RESPONSE TO GROUP B STREPTOCOCCUS. Madeline Ambrus Lillie, Carolyn S. Cody, Steven D. Douglas, Richard A. Polin. (Spon. by W.W. Fox). Div. of Neonatology and Allergy-Immunology, Children's Hospital of Phila., Univ. of Pa. School of Medicine, Philadelphia, PA.

Group B streptococcal (GBS) infection is a major cause of neonatal mortality and morbidity. Host factors responsible for immunity to GBS include specific antibody, functional neutrophils and intact complement pathways. The role of cell-mediated immunity is unknown. To investigate the relationship between humoral immunity and cell-mediated immunity to GBS, the in vitro lymphocyte response to whole bacteria was studied in 13 healthy adult volunteers. Lymphocytes were isolated and cultured in 15% autologous serum in microtiter plates. Each well contained 2 X 10^6 cells and either 1) washed formalin killed type III GBS in cell to organism ratios of 1:10, 1:1, or 1:0.1, 2) PHA, or 3) SK:SD. ^{3}H thymidine incorporation was measured at 5,6 or 7 days. The serum concentration of type III GBS antibody was determined with an enzyme immunoassay. Six of 13 adults responded to type III GBS as defined by a stimulation index of >10 or absolute count >4000 CPM. The mean serum concentration of type III GBS antibody in responders (15.5±7.4 M± SEM ug/ml) did not differ significantly from non-responders (6.0±10.1 ug/ml). Maximal response to GBS occurred at a cell/organism ratio of 1:1, ($p<.05$). Thymidine incorporation was significantly greater at 6 days than at 5 or 7 days ($p<.005$). The in vitro lymphocyte response to type III GBS may be an important determinant of host susceptibility to infection.

1040 ENUMERATION OF GROUP B STREPTOCOCCI (GBS) IN COLONIZED INFANTS. Daniel V. Lim, Mary H. Marnell and Keith S. Kanarek (spon. by Lewis A. Barness) Department of Biology, University of South Florida and Department of Pediatrics, University of South Florida College of Medicine, Tampa, Florida.

It is unknown why only 1 in 100 colonized infants develop GBS disease. There have been several semiquantitative studies of colonization in mothers, but no quantitative studies on magnitude of colonization in infants. This study was designed to determine if infants symptomatic for early-onset GBS disease (respiratory distress) are heavily colonized with the bacteria. Newborn infants suspected of early-onset disease at Tampa General Hospital were cultured at three or more sites with Culturette^R dual swabs. The swabs were used for identification of GBS by slide co-agglutination and enumeration of the bacteria by viable plate count. Eleven (13%) of 85 infants cultured were found to be colonized with GBS at one or more sites. Two of these eleven colonized infants were symptomatic for early-onset GBS disease. Both symptomatic infants were heavily colonized ($\geq 2.9 \times 10^3$ bacteria/swab) with GBS of serotype I. In comparison, only one of the nine asymptomatic infants was colonized with GBS of serotype I and the colonization was of a low magnitude ($<2.0 \times 10^1$ bacteria/swab). The other eight infants were colonized with nontype I GBS, generally at low numbers. The two asymptomatic infants that were heavily colonized with GBS were colonized with bacteria of serotype III. These preliminary data suggest a possible correlation between heavy colonization of neonates with specific serotypes of GBS and the development of early-onset group B disease.

● 1041 EPIDEMIOLOGY OF *HAEMOPHILUS INFLUENZAE* DISEASE DETERMINED BY OUTER MEMBRANE PROTEIN COMPOSITION. Marilyn R. Loeb, Richard A. Insel, and David H. Smith, University of Rochester Medical Center, Department of Pediatrics, Rochester, New York.

The outer membrane protein composition of 50 disease isolates of *H. influenzae*, including strains of all 6 serotypes as well as untypable strains, has been determined by SDS-PAGE. Results show the existence of considerable strain variation in the mobility of some, but not all, of the proteins. For example, the 28 type b strains examined could be classified into 8 subtypes according to differences in the mobilities of 4 of the 7 major outer membrane proteins. Untypable strains showed even greater diversity. Using these differences as an epidemiological tool, we have found the following: (1) A recent outbreak of type b meningitis involving 17 Rochester area children over a 6-week period was caused by at least 5 different strains, indicating that a single "killer" strain was not responsible for the epidemic. (2) In all 7 patients examined, type b isolates from different anatomic sites in the same patient were identical, suggesting that a single bacterium initiated the disease. (3) Isolates from a patient experiencing a repeat type b infection were identical, suggesting that the same strain was responsible for reinfection. In addition to these findings we have also discovered: (1) Some outer membrane proteins are common to all strains. (2) Children recovering from systemic type b disease raise serum antibodies to outer membrane proteins. Studies are currently in progress to identify the particular immunogenic proteins.

1042 HEMOPHILUS INFLUENZAE BIOTYPE PREDICTS VIRULENCE. Sarah S. Long, Mary J. Teter (Spon. by A.M. DiGeorge) Temple Univ. School of Medicine, St. Christopher's Hospital for Children, Department of Pediatrics, Philadelphia.

Biotype and serotype were determined for 359 isolates of H. influenzae(Hi) to define relation to virulence and antibiotic resistance. Of 61 patients with Hi isolates from invasive sites 95% were serotypable(S+) and 79% were biotype(B)I. 87% of cases of Hi meningitis, cellulitis, bacteremia and arthritis were caused by BI;50% of epiglottitis and pneumonia were. All 71 isolates from inflamed conjunctivae were not serotypable(S-);83% were B II or III;only 8% were BI. Frequency and biotype of conjunctival Hi did not differ with age or season. 147 children with Hi respiratory isolates were grouped according to illness and probable pathogenic role of Hi. 90% of respiratory isolates were S-. Group 1(well or ill, Hi not related)had no S+ isolates and only 7% were BI. Group 2(ill, Hi possibly/probably related)had significantly more S+(17%)and BI(35%). Group 3(cystic fibrosis patients)also had significantly more S+(23%)and BI(45%). Ampicillin resistance of respiratory isolates was similar for all groups. Otitis media(Hi middle ear isolate)was significantly associated with BI(8/13,61%) but not S+(1/13). Ampicillin resistance of Hi(any source-299 patients)was greater in S-(18%)than in S+(10%)strains. Resistance varied significantly with biotype;(BI highest(46%) and BI lowest (10%). In summary serotype positive Hi was associated with invasive disease. Biotype I Hi(serotype positive and negative)was associated with invasive disease, otitis media, and cystic fibrosis but had least resistance to ampicillin.

Abstract Withdrawn

1044 LOCAL PROTECTION OF THE MIDDLE EAR DURING RECURRENT OTITIS MEDIA. Gabriel Marshak, William J. Doyle, John S. Supance, and Erdem I. Cantekin (Spon. by Richard H. Michaels) University of Pittsburgh School of Medicine, Dept. of Otolaryngology, Children's Hospital of Pittsburgh, Pittsburgh.

A series of longitudinal studies of otitis media secondary to three strains of bacterial pathogens were conducted in the chinchilla animal model in order to define the importance of local and systemic mechanisms for the protection of the middle ear. The three strains of bacteria used were: Streptococcus pneumoniae type 6A and type 7F, and β-lactamase producing non-typable Haemophilus influenzae. Initially, the bacteria were inoculated unilaterally into the epitympanic bulla. Following the resolution of the induced otitis media with effusion, bacterial challenges were repeated ipsilaterally and contralaterally. Middle ear condition was assessed employing otomicroscopy, tympanometry and periodic direct inspection and culture. The results for the three strains of bacteria were similar. The ipsilateral middle ear was resistant to subsequent infection whereas the contralateral middle ear evidenced a disease course similar to that induced by the initial challenge. Thus resistance to subsequent infection appears to be a local phenomenon with little systemic participation.