799 QUANTITATION OF BACTEREMIA IN CHILDHOOD AND ITS RELATIONSHIP TO SEPTIC COMPLICATIONS Mathuram Santosham and E. Richard Moxon (Spon. by John Neff), Department of Pediatrics, Baltimore City Hospital and Johns Hopkins University, Baltimore, Maryland

The magnitude of bacteremia may be a primary determinant of whether children develop septic complications, such as meningitis. Therefore, quantitative blood cultures (QC) were sought in 243 febrile children from whom routine blood cultures (RC) were obtained because of unexplained fever. QC were obtained as follows; when blood was obtained for the RC, an additional 0.5 ml blood was drawn into a sterile syringe rinsed with polyanethol sulfonate; 10 and 100 μl was plated directly on to 1)sheep blood agar and 2)brain-heart infusion media enriched with Levinthal base. There were 14 positive QC from 12 patients. These were 7 Hemophilus influenzae b, 5 S.pneumoniae and 2 S. aureus. Bacterial counts ranged from 20->10^4 per ml. In three cases of H.influenzae b meningitis, bacteremia exceeded 10^3 organisms/ml. In nine other patients with bacteremia unassociated with meningitis, counts were 10^2-10^3/ml in three (pneumonia, peritonitis, isolated bacteremia) and <10^2/ml in six (one pneumonia, one epiglottitis, four isolated bacteremia). Bacteremia detected by QC was identified as early or earlier than its recognition by RC. In three patients, bacteremia was detected by QC but not by RC. Quantitative cultures aided in the early detection of bacteremia and provided data that may help in identifying those children at highest risk for developing septic complications such as meningitis.

800 COMPARISON OF ANTIGEN DETECTION METHODS IN A PRIMATE MODEL OF HEMOPHILUS MENINGITIS. D.W. Scheifele, R. Daum, V. Syriopoulou, G. Siber and A. Smith, Children's Hospital Medical Center, Dept. of Med., Boston, Mass.

An infant primate model of H. influenzae b (HIB) infection was employed to compare the sensitivity of two rapid methods to detect infection. Latex particle agglutination (LPA) and CIE assays developed in this laboratory detect 0.5 and 1.0 ng/ml of HIB antigen, respectively. Serial quantitative cultures were performed on 19 monkeys with bacteremia and meningitis and corresponding samples tested for antigen. Both assays detected antigen in the serum of all 19 bacteremic animals, with detection a function of duration and density of infection. LPA became positive earlier:70% of animals were LPA^+ after 8 hrs of bacteremia and 100% after 36 hrs, compared with 10% and 60% using CIE. After 80 hrs, 84% of sera were CIE^+. LPA detected fewer bacteria in blood:after 36 hrs of constant-density bacteremia, 6/6 animals with 10^2 - 10^3 cfu/ml were LPA^+ while 1/6 was CIE^+. CIE became reliable at bacterial concentrations $\geq 10^4$/ml of blood; at 36 hrs, 8/9 such samples were CIE^+. CSF was obtained from 18 monkeys within 24 hrs of meningitis onset (mean bacterial conc. 1.4×10^4 cfu/ml CSF). LPA was positive in 15/17 samples, detecting 4/6 containing 10^2 cfu/ml and all 11 with $\geq 10^3$/ml. CIE was positive in 9/18 samples, detecting 3/12 containing 10^2 - 10^3 cfu/ml but 6/6 with $\geq 10^4$/ml. 24 hrs later, LPA was positive in all CSF samples and CIE in 16/18 (84%). We conclude that LPA is superior to CIE, detecting antigen earlier and with fewer bacteria (10^2 - 10^3 bacteria/ml of serum or CSF).

801 COUNTERCURRENT IMMUNOELECTROPHORESIS (CIE) IN GROUP B STREPTOCOCCAL DISEASE. Penelope G. Shackelford, Barbara W. Stechenberg (Spon. by Ralph D. Feigin), Washington Univ. Medical School, St. Louis Children's Hospital, Dept. of Pediatrics, St. Louis.

CIE was utilized for the detection of type and group specific antigens in the body fluids of 38 infants with Group B streptococcal infection. Serum, CSF, or urine from 27 of 38 (71%) contained detectable type or group specific antigen. Serum was positive by CIE in 10 of 24 patients tested and 7 of these 10 infants died (70%). Only 2 of 14 (14%) infants without detectable antigen in their serum died ($p<.05$). Antigen was detected in the CSF from 7 of 10 infants tested and none of these patients died. Antigen was detectable by CIE in 14 of 15 (93%) of urine samples which had been concentrated using an Amicon® filter. In 9 instances, samples of urine, serum and CSF were available from the same patient and 8 had antigen detected in one or more body fluids. Urine was positive in all 8 patients and in 3 instances, was the only positive fluid. Of 27 samples with detectable antigen,10 were positive using both group and type specific antisera, 10 were positive only with the type specific antiserum and 7 were positive only with the grouping antiserum. The type antigen detected in the CSF or serum always corresponded to the Lancefield type of the infecting organism. Some cross reactions occurred when testing concentrated urine but the type specific reaction corresponding to the organisms cultured was always strongest. The detection of Group B Streptococcal antigens in the body fluids by CIE can be performed rapidly and provides useful diagnostic information.

802 ROLE OF ANTIBODY AND THE CLASSICAL AND ALTERNATIVE COMPLEMENT PATHWAYS ON THE OPSONIZATION OF GROUP B STREPTOCOCCI. Ann O. Shigeoka, Robert T. Hall, Val G. Hemming, and Harry R. Hill, Dept. of Pediatrics and Pathology, Univ. of Utah, Salt Lake City, and Children's Mercy Hospital, Kansas City, Missouri.

In previous studies we have shown that neonates who develop Group B streptococcal sepsis usually lack opsonins to their infecting strain. This investigation was designed to determine the role of antibody and complement in opsonization of these organisms. We also evaluated the effects of fresh blood transfusions on serum opsonic activity in neonates. Group B streptococcal opsonins were determined by a chemiluminescence procedure previously described (JCI 58:1379, 1976). Opsonic activity for types Ia and II was significantly depressed by heating and by inactivation of the classical pathway with Mg^{++}EGTA. Similar treatment of fresh serum containing type III antibody did not alter opsonic activity for this strain. In the absence of heat stable antibody, fresh serum demonstrated no alternative pathway opsonic activity for group B streptococcal strains; factor B was not activated by electrophoretic analysis; and C3 was not deposited on the cell surface. Comparison of pre and post transfusion sera from 19 neonates showed a rise in opsonic activity only when heat stable antibody was present in donor serum. These results confirm the importance of heat stable antibody and the classical pathway in opsonization of Group B streptococci but do not indicate a major role for the alternative pathway. Therapy aimed at supplying passive immunity to infected neonates will require the presence of heat stable opsonins.

803 SIMULATION OF RHEUMATIC RESPONSE TO STREPTOCOCCAL CARBOHYDRATE IN RABBITS BY USE OF ADJUVANT. Stanford T. Shulman and Elia M. Ayoub, Univ. of Florida, Dept. of Pediatrics, Gainesville.

Previous studies from this laboratory demonstrated that patients with inactive rheumatic valvular disease generally maintain elevated levels of antibody to Group A streptococcal carbohydrate (A-antibody) following acute rheumatic fever, while A-antibody levels fall to normal in other post-streptococcal states. Immunization of rabbits with killed Group A streptococci was performed to define the conditions necessary to produce sustained A-antibody. A-antibody was assayed by the Farr technique utilizing ^{14}C-A-carbohydrate. Rabbits immunized by Lancefield's schedule of thrice-weekly IV injections for 4 weeks produced very high A-antibody levels which promptly fell; boosting with a similar schedule induced the same response. Transient low A-antibody levels followed a single IV immunization, peaking at 6-8 days and falling by 14-20 days. Single IV boosts consistently recalled transient A-antibody responses. In contrast, a single subcutaneous (SC) dose of vaccine in complete Freund's adjuvant (CFA) led to sustained high A-antibody levels. Peak levels were reached by 35-45 days and remained elevated for at least 120-185 days. SC vaccine without CFA induced low level A-antibody which declined promptly and disappeared by 50 days. Because adjuvant effect appears to correlate with slow antigenic release from the depot, these data support the possibility that the sustained serum A-antibody levels in rheumatic heart disease represent chronic antigenic stimulation by streptococcal A-carbohydrate or by cross-reactive tissue antigens.

804 BACTERICIDAL ANTIBODY TO NON-TYPABLE STRAINS OF HEMOPHILUS INFLUENZAE. Paul A. Shurin, Stephen I. Pelton, Ira B. Tager and Dennis L. Kasper (Spon. by J.O. Klein). Harvard Medical School, Boston University School of Medicine, Boston City Hospital, Depts. of Pediatrics and Medicine, Boston.

Ninety per cent of H. influenzae from middle ear effusions are non-typable (HiNT). We have used a bactericidal antibody assay to classify HiNT and detect specific antibody. Rabbit antisera to two strains (T1 and T2), with complement, kill homologous organisms. Anti-T2 is specific after absorption with T1 bacteria(anti-T2 abs). Sensitivity to these sera distinguished 4 serogroups among 22 HiNT strains from middle ear effusion:

Group	Anti-T1	Anti-T2	Anti-T2 abs	# of Strains
T1	+	+	-	8
T2	+ or -	+	+	6
T2x	-	+	-	3
SR	-	-	-	5

Human sera were tested for activity against T1, T2, an SR strain, and a type b strain:

	PER CENT WITH BACTERICIDAL ANTIBODY Age Group (# tested)			
	4-36 months(25)	5-9 yrs(37)	22-53 yrs(17)	Cord Sera(20)
T1	84.0	100	100	100
T2	52.0	91.9	88.2	90.0
SR	4.0	5.4	11.8	20.0
b	4.0	27.0	29.4	25.0

The age and strain differences in prevalence of antibody demonstrate the specificity of this typing scheme. Classification of HiNT should facilitate epidemiologic study of otitis media.