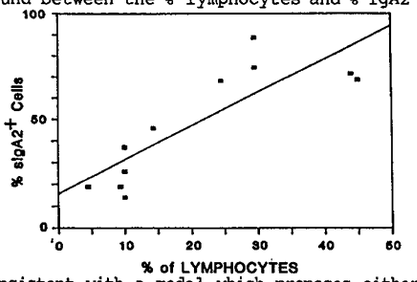
1027 **Association of IgA Subclass Concentration and Lymphocyte Subpopulations in Human Colostrum** H.B. Slade, S.A. Schwartz, Ann Arbor, MI.

The mechanisms regulating expression of IgA subclasses are poorly understood. While adult serum contains ~90% IgA1 and ~10% IgA2, breast milk contains a 40/60 mix respectively. In the present study colostrum was collected from 11 donors (days 1-5 post-partum), the cells washed through a fetal calf serum cushion, separated over ficoll-Hypaque, and twice washed with buffer. The mononuclear cell fraction was assessed by Wright-Giemsa stain and flow cytometry using fluorescein-conjugated monoclonal anti-T4, T8, IgA1 and IgA2 antibodies. A strong correlation was found between the % lymphocytes and % $IgA2^+$ cells (R=.842).

The T4/T8 ratio did not correlate, and averaged 1.11+/-.67

No correlation was found with surface $IgA1^+$ cells.



The findings are consistent with a model which proposes either recruitment to or expansion of $sIgA2^+$ lymphocytes in colostrum.

●1028 FUNCTIONAL ANALYSIS OF IgA PRODUCING CELLS FROM PATIENTS WITH SELECTIVE IgA DEFICIENCY. Leonard D. Stein, Chaim M. Roifman, Sasson Levi, Erwin W. Gelfand, Marcia A. Chan and Hans-Michael Dosch. Research Inst. The Hospital for Sick Children, Div. of Immunology, Toronto, Ontario, Canada.

IgA deficiency is the most common humoral immune defect. We examined blood lymphocytes from 4 patients with Ataxia Telangiectasia and 6 with common IgA deficiency. All had absent serum IgA but .5-2% $sIgA^+$ circulating B cells. PWM and EBV were used for the stimulation of these cells either in high density cultures (10^6 cells/well) or in low density, limiting dilution cultures (1-10^4 cells/well), which restrict cell:cell interactions. IgG, A and M secreted into culture supernates was measured by RIA and ELISA after 8 d (PWM) or 4 wk of culture (EBV). Cells from patients failed to produce IgA in high density cultures. In EBV stimulated limiting dilution cultures, IgA producing cells were detected in all patients. The frequency of IgA-committed, EBV transformable cells was normal in patients with uncomplicated IgA deficiency (approximately 1/100 B cells). However, in patients with AT, the frequency of IgA producing cells was up to 40 times lower. Our experiments demonstrate the presence of IgA producing cells in all patients with IgA deficiency studied. No evidence for IgA specific B cell defects could be delineated in common IgA deficiency. In AT, abnormalities of B cell differentiation may lead to the development of a smaller, IgA-committed B cell compartment.

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1029 PLACENTAL CELLS OF FETAL ORIGIN SYNTHESIZE GAMMA INTERFERON. E.R. Stiehm, D. Murakami, T. Chin, A. Goldsobel, B. Ank, J. Giorgi and C. Spina. UCLA Dept. of Pediatrics, Los Angeles, CA.

The placenta is a rich source of immunocompetent mononuclear cells (MNCs) that have microbicidal, cytotoxic and lymphokine-producing capabilities (interleukin-2 (IL-2)). We have identified several immunologic properties of placental cells that differentiate them from the corresponding neonatal cord blood cells. Previous studies have established that cord blood MNCs have deficient natural killer (NK) activity and gamma interferon (IFN-γ) production. The placental MNCs (prepared by collagenase and DNAase digestion followed by Ficoll-Hypaque density gradient separation) are of fetal origin, as shown by chromosome analyses of cells from 4 placentas from male infants, all of which were XY. Cell sorting indicates a significantly higher proportion of NK Leu 11 cells in placenta compared to the corresponding cord blood (24 $\pm$ 4% vs 15 + 4%). All supernatants from placental MNC samples incubated with PHA for 48 hours produced significant (>10 IU/ml) amounts of γ-IFN, (mean 27 $\pm$ 5 SEM) as assayed by a new radio-immunoassay (Centocor). This assay correlates well with the standard bioassay. Most (19/21) PHA-stimulated cord blood MNCs showed deficient production (<4 IU/ml) of γ-IFN. This could be partly corrected by incubation with IL-2 or IL-2 + PHA in 6/11 cord bloods. 22 adult PHA-stimulated MNC produced large amounts of γ-IFN (51$\pm$17 IU/ml).

These properties of placental immune cells suggest a separate and unique intrauterine immune system that preceeds the dormant fetal immune system. The latter may be activated by the use of interleukin 2.

1030 PRETRANSLATIONAL REGULATION OF THE SYNTHESIS OF THE THIRD COMPONENT OF COMPLEMENT (C3) IN HUMAN BLOOD MONOCYTES BY THE LIPID A PORTION OF LIPOPOLYSACCHARIDE (LPS). R.C. Strunk, F.S. Cole. Depts. of Pediatrics, University of Colorado Medical School, National Jewish Hospital and Research Center, Denver, Colo., and Harvard Medical School, The Children's Hospital, Boston, MA. 02115

LPS isolated from the outer membrane of gram negative bacteria has been shown to increase specifically the production of C3 by human blood monocytes. The availability of cDNA probes for C3 as well as C2 and factor B and structurally altered LPS permitted examination of the biosynthetic regulation of and structural requirement for this effect of LPS. Poly-A RNA was isolated from monocytes incubated in the presence and absence of LPS and subjected to Northern blot analysis using single strand (M13) cDNA probes. C3 mRNA increased from 5 to 10 fold without a comparable increase in C2 or factor B mRNA. To determine the structurally relevant portions of the LPS molecule, monocyte monolayers were incubated in the presence of lipid A inactivated (by alkaline hydrolysis) LPS, LPS isolated from a polysaccharide deficient mutant of S. minnesota, lipid X, and a monoacylglucosamine-1-phosphate derivative of lipid X. Using biosynthetic labelling with 35S methionine, immunoprecipitation, and SDS-PAGE, polysaccharide deficient LPS and lipid X increased newly synthesized C3, but lipid A inactivated LPS and the lipid X derivative did not. These data suggest that LPS increases C3 synthesis by human monocytes pretranslationally and that the reducing end subunit of the lipid A portion of the molecule is the minimal structural requirement for this activity.

† 1031 HEMOPHILIAC IMMUNODEFICIENCY: INFLUENCE OF EXPOSURE TO FACTOR VIII, HUMAN T LEUKEMIA VIRUS (HTLV III) AND HERPESVIRUSES. JL Sullivan, FE Brewster, DB Brettler, AD Forsberg, PH Levine. Univ. of Mass. Med. Sch., Worcester, MA, Dept. Ped. Med.

We have evaluated 121 patients with hemophilia A for cellular immune defects and exposure to HTLV III, Epstein-Barr Virus (EBV) and cytomegalovirus (CMV). Immunoregulatory defects were found in the majority of the patients studied: decreased OKT.4/T.8 ratio, decreased lymphocyte proliferation in response to phytohemagglutinin (PHA), anti-immunoglobulin, pokeweed mitogen (PWM) and streptolysin O, and decreased natural killer (NK) cell activity($p^<.05$). Delayed hypersensitivity skin test responses to a battery of 6 antigens were completely absent in 42% of patients. 70% of the population had antibodies to HTLV III, EBV and 43% to CMV. Multivariate analysis was performed at a confidence level of $p\leq.01$ and demonstrated that the single most important predictor of immunoregulatory defects was the amount of Factor VIII used. Absolute numbers of T helper cells, helper/suppressor cell ratios, PHA and PWM responses were all negatively correlated with units/kg of Factor VIII utilized. Out of 10 correlations found between the presence of viral antibodies and immune abnormalities, 7 involved HTLV III alone or in combination with CMV and/or EBV; notably positive correlations with PHA, anti-immunoglobulin, T-suppressor cells. EBV correlated negatively with T-suppressor cell populations, while correlations with CMV occurred only in the presence of antibodies to EBV and/or HTLV III. These studies demonstrate that treatment of hemophilia with Factor VIII concentrate is associated with long-term immunoregulatory defects and that exposure to HTLV III plays a role in producing further defects.

† 1032 COMPLETE ABSENCE OF NATURAL KILLER (NK) CELLS ASSOCIATED WITH NEAR FATAL VARICELLA INFECTION. JL Sullivan, KS Byron, CA Biron. Univ. of Mass. Med. Sch., Worcester, MA, Dept. of Ped. and Path.

A 13 year old female presented with overwhelming varicella infection with varicella pneumonia. Immunological evaluation revealed complete absence of natural killer cell activity with normal T lymphocyte subsets and proliferative responses. The varicella infection was controlled with a course of acyclovir and the patient recovered with severe scarring. Immunological studies following convalescence have revealed a persistent lymphopenia (564$\pm$205), normal quantitative immunoglobulins and T lymphocyte proliferative responses. Nucleoside phosphorylase levels were normal. Neutrophil chemotaxis was normal and mononuclear cells reacting with the OKM-1 monoclonal were present. NK cells in the peripheral blood enumerated by the monoclonal antibody Leu 11 were markedly deficient (1-5%). Complete absence of NK cell activity (50:1 E/T ratio) against K562 (0.5$\pm$0.8 specific immune release, $p^<.0001$) has persisted. In vitro NK cell function was completely unresponsive to recombinant γ interferon and IL-2. Mixed lymphocyte culture induced T cell mediated cytotoxicity against lymphoid tumor targets was normal. No Leu 11 positive cells could be induced to proliferate in response to K562 and lymphokine (IL-2) activated killing was absent. Percoll gradient enrichment for NK cells revealed complete absence of Leu 11 positive large granular lymphocytes. Percoll enriched populations failed to form normal conjugates with K562 and killing in agarose was absent. These results suggest that NK cells play an important role in the early immune response to varicella virus.