715 VARIABLE EXPRESSION OF "X-LINKED INFANTILE AGAMMAGLOBULINEMIA" (X-LA). Hans D. Ochs, John L. Sullivan, and Ralph J. Wedgwood. Department of Pediatrics, University of Washington, Seattle.

X-LA is a syndrome of males, characterized by early onset of recurrent pyogenic infections, markedly diminished or undetectable serum immunoglobulins, depressed or absent humoral immune responses, intact cellular immunity, and lymphoid tissues devoid of plasma cells, germinal centers, and follicle formation. Rigorous criteria include other affected male maternal relatives.

We have evaluated 11 boys with presumptive X-LA, from 6 unrelated families. Three families each had only one affected individual, although fulfilling the criteria in all other respects. Three families had at least one affected nephew maternal-uncle pair. In one of these, both affected relatives had no B-cell function. In the other two, the uncles had classic X-LA, including failure to clear antigen (ØX 174) or make antibody, and lymphoblastoid cell lines (LCL) could not be induced with Epstein-Barr virus. The affected nephews, however, had typical variable immunodeficiency: immunoglobulins were low, not absent; circulating B-lymphocytes were present (in decreased numbers), and a low but measurable antibody response, including memory was demonstrable. In one nephew an LCL with male karyotype and B-cell characteristics was established. Mothers of the affected males were immunologically normal. These findings demonstrate a variable expression of the B-cell defect in X-LA. Indeed since sex-linked inheritance remains unproven, perhaps other mechanisms of pathogenesis, inheritance and expression deserve consideration.

716 HUMAN LYMPHOCYTE TRANSFORMATION FOLLOWING INHIBITION OF PURINE NUCLEOSIDE PHOSPHORYLASE. Ochs U, Osborne W, Chen S-H, Scott CR. University of Washington, Department of Pediatrics, Seattle, Washington.

The genetic absence of purine nucleoside phosphorylase (NP) is associated with an isolated T-cell immune deficiency in humans. NP is responsible for the conversion of inosine to hypoxanthine and guanosine to guanine. We have synthesized a non-metabolizable compound, 6-hydroxy-9-p-aminobenzylpurine (HABP), an inhibitor of NP that has a Ki of 250 μM. The presence of HABP reduced the incorporation of ^{3}H-thymidine into DNA in phytohemagglutinin stimulated human lymphocytes from healthy volunteers to 30% of control values. Patients with NP deficiency have increased levels of inosine and guanosine in their plasma. The addition of inosine to the culture medium of stimulated lymphocytes at 4 mM had no inhibitory effect on DNA synthesis as measured by thymidine incorporation. The addition of 2 mM guanosine, however, reduced thymidine incorporation to 4% of control lymphocytes. The inhibitory effect of guanosine could be partially corrected by the addition of hypoxanthine to the medium at 500 μM concentrations. The addition of uridine to the medium at concentrations to 1 mM did not reverse the inhibition induced by guanosine. These studies indicate that the T-cell abnormality observed in NP deficiency can be mimicked by two related mechanisms, 1) by inhibiting NP in normal lymphocytes, or 2) by increased concentration of guanosine in the culture medium.

717 THE EFFECT OF NORMAL HUMAN SERA (NHS) ON COMPLEMENT CONSUMPTION (CC) BY IMMUNE SERUM GLOBULIN (ISG) OR ISOLATED HUMAN IgG (IgG). Lauren M. Pachman, Margaret D. Mikus, Sandra M. Baldwin Northwestern Univ. Med. Sch., Children's Mem. Hosp., Dept. of Ped., Chicago.

We reported (Ped Res 9 Abst #459,1975) that CC was correlated with disease activity in juvenile rheumatoid arthritis ($p<.001$). This assay heat inactivates (HI) the endogenous complement of the test sera: 56°C x 30'. The effect of heating ISG (Cohn fraction II) or DEAE-Sephadex purified IgG was next studied. It was found that: 1) in the absence of NHS, unheated IgG (initial concentration 15.4mg/ml) had essentially no CC. However, ≥.5mg/ml unheated deaggregated ISG had increasing CC. 2) heating concentrated IgG without NHS resulted in more CC than heating diluted solutions. 3) addition of NHS prior to heating either ISG or IgG greatly decreased CC: NHS and up to 15.4mg/ml IgG had no CC while increasing CC was seen with 1.5mg/ml or more of ISG. 4) addition of HI NHS to heated IgG or ISG prevented CC to a lesser extent than when NHS was heated with added immunoglobulin. It is concluded that the CC assay is not appreciably affected by HI of normal sera containing a total amount of up to 23mg/ml of IgG. Supported by grants from the Nat'l. Fdn. and the Ill. Chap. Arth. Fdn.

718 CHARACTERISTICS OF IMMUNE RESPONSE TO VARICELLA INFECTION IN NORMAL AND IMMUNOSUPPRESSED HOST. Pratibha Patel, Shams Yoonessi, Anne Gershon, Yasuo Chiba, and Pearay L. Ogra, State Univ. of N.Y. at Buffalo and N.Y. Univ. Med. Center, N.Y.

Employing the techniques of indirect fluorescent antibody staining (IFA), e-rosette formation and in-vitro lymphoproliferative responses (LTF) to phytohemagglutinin (PHA) and inactivated varicella-zoster virus antigen (V-Z), the development of antibody and cell-mediated immunity (CMI) was studied, following naturally acquired clinical varicella in groups of normal immunologically competent children and in patients with leukemia on intensive immunosuppressive therapy. The immune response in normal children was characterized by the detection of significant antibody and CMI response 10-14 days after the onset of rash. The CMI activity gradually declined, when tested sequentially over the period of 110 days. The antibody titers manifested little or no change. All patients elicited significant responses to PHA and no change was observed on subsequent follow-up. Immunosuppressed children who recovered uneventfully after varicella, manifested specific immune response to V-Z which was similar to that observed in normal children. On the other hand, the CMI to V-Z in immunosuppressed subjects with fatal varicella infection was characterized by conspicuous absence of CMI activity to V-Z, 7-10 days after the onset of rash. These observations suggest that varicella infection induces CMI response in normal subjects and the development of such response may determine the outcome of V-Z infection in immunosuppressed individuals.

719 NORMAL AND ABERRANT B-CELL DIFFERENTIATION IN BONE MARROW. Elliott R. Pearl, Larry B. Vogler, William M. Crist, Alexander R. Lawton, Max D. Cooper. Univ. of Alabama in Birmingham, Children's Hospital, Departments of Pediatrics and Microbiology, Birmingham, Alabama.

The first member of the B-cell series is a large lymphoid cell (pre-B) with cytoplasmic but undetectable surface(s) IgM. Pre-B and sIg+ cells were examined in marrow and blood from 6 normals and 18 patients with Ig deficiencies or blood disorders. Of normal nucleated marrow cells, 0.6±0.5% were pre-B and 2.1±1.4% were sIgM+ B cells. 9 boys with infantile agammaglobulinemia were deficient in sIgM+ cells in blood and marrow but had normal proportions of marrow pre-B cells (0.6±0.5%). Marrow-cell cultures from 2 of these boys failed to generate B cells with or without addition of theophylline or dibutyryl cAMP. Both pre-B and B cells were absent in marrow from 2 men with thymoma and hypogammaglobulinemia; both also lacked eosinophils. A normal frequency of pre-B cells was found in 5 hypogammaglobulinemic patients with normal circulating B cells. Frequency of pre-B cells was increased (4.0 and 4.6%) in 2 boys' marrow after histocompatible marrow transplantation for aplastic anemia or leukemia. Pre-B cells were not found in blood(7), spleen(2), or lymph nodes(2). Our results (i) strengthen the idea that B cells are derived from sIg- pre-B cells resident in bone marrow, (ii) indicate that pre-B to B-cell conversion is consistently defective in infantile agammaglobulinemia, (iii) provide more evidence for defective stem cell differentiation in immunodeficiency associated with thymoma, and (iv) suggest that regulation of B-cell generation in bone marrow can be experimentally modulated in man.

720 RAPID DETERMINATION OF IgM USING LATEX REAGENT IN NEONATES. Alistair G. S. Philip, Jean Hewitt (Spon. by Jerold F. Lucey). University of Vermont College of Medicine, Department of Pediatrics, Burlington.

Chronic intrauterine infection commonly results in increased levels of immunoglobulin M (IgM). It is recognized that elevated levels of IgM are non-specific, but useful as a screening test. This is particularly true in infants with intrauterine growth retardation. In many hospitals IgM testing is done infrequently (e.g., twice weekly). The use of a rapid method for determination was tested for reliability by doing paired determinations of IgM by a latex method and an immunoelectrophoretic method. Samples were obtained from babies being investigated for possible bacterial or viral infection. The "positive" on the latex method is currently set at 30 mg% or more. The results were:

Immuno-IgM	Latex IgM	
	Negative	Positive
≤ 30 mg%	147	3
> 30 mg%	5	19

When the false negatives (5) and false positives (3) were examined, only 1 false positive fell outside the ± 5 mg% range and was within 10 mg%. Introduction of another reagent set at 20 mg% could eliminate this error, in conjunction with the present reagent. The latex IgM method is easily performed, rapid (within 10 minutes) and inexpensive. A positive test could influence further investigations (e.g., viral cultures, long-bone x-rays, etc.).