patients are Medicaid eligible. 44% are of Puerto Rican or other Latin American origin. 5% could be termed "hippies". 45% are boys. A review of records of 523 patients registered in a 16 month period shows that 31% are known to be seriously involved with drugs other than marijuana. These are patients not referred to our service for a drug problem, but, for various medical conditions or routine examinations. 19% of our patients use heroin; 9% use barbiturates and other depressants; 7% use amphetamine and other stimulant pills ("ups"); 6% use LSD or mescaline; 5% use methedrine by injection ("speed"). Mixed use is common. 37% of these drug abusers are 16 years of age or younger. 60% are boys. 36% of the drug users are of Puerto Rican or other Latin American origin. 13% are "hippies". The majority live at home and attend school. Information concerning patterns of use, means of acquiring drugs, attitudes and knowledge of effects, based on in-depth interviews with about 40 drug users, will be discussed.

IMMUNOLOGY

HL-A phenotypes in leukemias: A family study. SUSIE W. FONG, ROBERT LUNDAK, and SHARON BRITT (Intr. by Thos. L. Nelson). *Univ. of Calif., Irvine, Calif.*

The acceptance or rejection of leukemic cells by the host may be related to transplantation immunology. Viral infections of cells are known to alter the antigenic composition of cell membranes and lead to the development of new antigens, deletions with replacement by fetal forms, and/or partial development of specific antigens. Search for aberrant HL-A antigens in leukemias may lead to the discovery of tumor-specific antigens. HL-A phenotypes were determined by microcytotoxicity and absorption methods in 20 patients with acute lymphocytic leukemia (ALL) and 50 family members; 10 patients with chronic granulocytic leukemia; 20 patients with other lympho- and myelo-proliferative disorders and tumors. Seven of 16 ALL patients possessed one or two HL-A antigens not expected from family analysis. The aberrant HL-A specificities differed from patient to patient. Five of 16 ALL patients demonstrated three HL-A alleles per segregant group not found in family members. The gene frequency of HL-A7 in the ALL group was lower than expected ($p < 0.01$). There also appears to be a paucity of HL-A specificities at the locus of the second segregant group (5, 7, 8, 12) in ALL ($p < 0.01$). No relationship was found between certain HL-A genotypes and ALL. Malignant transformation of cells did lead to aberrant HL-A patterns. The discovery of HL-A specificities unique to the ALL patient through family analyses may form a basis for the development of immunotherapy.

The immunosuppressive effects of maternal plasma. SANFORD LEIKIN. *Children's Hosp. of D.C., and Geo. Washington Univ. Sch. of Med., Washington, D.C.*

Although the anatomical barrier which exists between the pregnant female and her fetus appears to be important, a modification of the immune response to histocompatibility differences may also play a role in the symbiosis of pregnancy. The one-way mixed lymphocyte reaction (MLC) expresses the reactivity of one population of cells against the histocompatibility antigens of another population. Therefore, the MLC was used to test the reactivity of newborns' and maternal lymphocytes to each other. It was found that, although maternal lymphocytes reacted to stimulation by cord blood cells, maternal plasma suppressed this response. It was also found that cord blood lymphocytes were hyporesponsive to stimulation by maternal cells as compared to adolescent-mother controls. This hyporeactivity was intensified in the presence of maternal plasma.

Further in vitro studies revealed that vaccinia to which the subjects had been previously immunized and suboptimal doses of phytohemagglutinin (PHA) stimulated pregnant females' lymphocytes significantly less well than adult males' cells, and that similarly stimulated cultures prepared with plasma from these females inhibited transformation of their own and male donors' lymphocytes. It appears, therefore, that maternal plasma contains factor(s) which inhibit(s) the MLC reaction, antigenic and PHA in vitro lymphocyte stimulation. This inhibitory effect of maternal plasma may be important in modifying the reactivity of maternal cells to fetal tissue. The inhibition of fetal lymphoid cells in the maternal circulation may also afford protection to the mother in a fetal graft-maternal host reaction.

Hormonal basis for sex differences in immunity. JEAN F. KENNY and JANET A. GRAY (Intr. by Richard H. Michaels). *Children's Hosp. of Pittsburgh, Pittsburgh, Pa.*

To investigate the greater susceptibility of the male to severe infections we have studied antibody production by individual spleen cells in immature and adult Swiss mice. Previous studies using the Jerne agar-plaque technique have shown: 1) following enteric colonization with *E. coli* or injection of small numbers of heat-killed *E. coli* (HKE) significantly more cells produce specific antibody in weanling and adult females than males; 2) after injection of small numbers of HKE (3×10^5) sexually mature females respond with significantly greater numbers of antibody producing cells (APC) than weanling females, but responses of adult males are only slightly better than those of male weanlings; 3) mean amounts of antibody produced by male and female cells are the same.

Responses of prepubertal ovariectomized females (OF) were compared to those of equal numbers of their sham-operated male (SM) and female (SF) littermates. Three weeks postoperatively, 4 days after intraperitoneal injection of 3×10^5 HKE, total APC/spleen ranged from 0–600. When individual totals of APC were ranked, responses of OF and SM were alike and those of SF were significantly better ($p < .001$). $1^0/_{28}$ SF vs. $^0/_{28}$ OF and $^7/_{30}$ SM were in the top third of the rank order. In a similar study total APC for castrated males and SM were the same. Estradiol-17β (500 ng) was given to 50 weanling males the week of challenge (5×10^5 HKE). In responses ranging from 0–1550 APC/spleen, the estradiol-treated males ranked higher than 53 saline-injected littermate controls ($p < .05$).

Findings show that the significantly better immunologic responsiveness of the female is dependent on ovarian function; small amounts of estradiol appear to enhance the proliferation of immunocompetent cells.

Further definition of two distinct types of congenital defects in plasma cell differentiation resulting in agammaglobulinemia. ALEXANDER R. LAWTON, DALE E. BOCKMAN, and MAX D. COOPER. *Univ. of Ala., Birmingham, Ala., and The Med. Coll. of Ohio at Toledo, Toledo, Ohio.*

Functional and morphologic evaluation of the immune system was carried out on 2 females with sporadic agammaglobulinemia (SA) dating from infancy and a male with X-linked agammaglobulinemia (XLA). All 3 patients had profound hypogammaglobulinemia, lacked isoagglutinins, and failed to synthesize antibodies to Salmonella H and O antigens following immunization.

All lacked plasma cells and cells exhibiting *cytoplasmic* fluorescence with fluorescine-labeled antibodies to IgM, IgG, and IgA in biopsies of ileum and stimulated lymph node. In contrast to the patient with XLA, patients with SA had prominent germinal centers in stimulated nodes; one had the syndrome of nodular lymphoid hyperplasia (NLH). Using criteria established for identification of bursa-derived lymphocytes in chickens, B-lymphocytes were identified by electron microscopy in the lymph node of one patient with SA; only T-lymphocytes were found in the patient with XLA. *Membrane-bound* immunoglobulins (IgM, IgG, and IgA) were detected by immunofluorescent studies of living peripheral blood lymphocytes of both patients with SA, but were not found on cells from the patient with XLA. The results indicate that in these patients with SA, lymphocytes of the plasma cell line are able to synthesize surface immunoglobulins and to respond to antigenic stimulation by germinal center formation but fail to differentiate to the stage of antibody secretion; lack of antibody-mediated "feedback" inhibition of antigen-induced proliferation may account for the NLH often observed in SA. Our observations also provide preliminary evidence that B and T lymphocytes can be distinguished at an ultrastructural level in man.

Phytohemagglutinin (PHA) skin test: A measure of intact cell-mediated immunity. RICHARD J. BONFORTE, R. MICHAEL BLAESE, MARCEL TOPILSKY, LOUIS E. SILTZBACH, and PHILIP R. GLADE. *Mt. Sinai Sch. Med., City Univ. N. Y., N. Y.*

PHA is a non-specific stimulant of normal lymphocytes resulting in vitro in their blast-like transformation with increased RNA and DNA synthesis, mitosis, and the release of putative mediators of cell-mediated immunity. Histologically the skin reaction to intradermal PHA shows perivascular infiltration of mononuclear cells, the hallmark of delayed-type hypersensitivity. Since prior exposure is not required—only the capacity to respond—PHA appears to be the ideal agent to assess cell-mediated immunity in vivo. In a continuing series of more than 30 individuals 2 μg of PHA intradermally produces erythema and induration in 24–48 hrs., despite the presence or absence of cutaneous reactivity to other skin-test antigens (PPD-S, histoplasmin, mumps, SK-SD, candida, and trichophyton). In a selected group of patients in vivo skin reaction to PHA correlates with in vitro response of the individual's peripheral blood lymphocytes to PHA. PHA in vivo thus appears to be an effective measure of intact cell-mediated immunity and is especially useful in infants and children who have had limited prior exposure to those antigens normally used as skin-test reagents.

Pernicious anemia, concordant twins, and immune dysfunction. ERWIN W. GELFAND, A. IZZET BERKEL, HERMAN A. GODWIN, ROSS E. ROCKLIN, JOHN R. DAVID, and FRED S. ROSEN. *Children's Hosp. Med. Ctr., Boston City Hosp., and Robert B. Brigham Hosp., Boston, Mass.*

The pathogenesis of pernicious anemia (PA) may be mediated by auto-immune mechanisms. However, the presence or absence of antibodies to intrinsic factor has not been clearly correlated with the clinical course of the disease. Three members of one family, a mother and identical twin daughters, were found to have classical PA and achlorhydria. Schillings tests revealed less than 1% excretion of the $Co^{57}B_{12}$ isotope. Repeat tests with orally administered intrinsic factor increased the isotope excretion to 10% or more. The twins, unlike their mother, had low levels of serum γG, absent γA, and normal γM globulins, and impaired antibody responses. The serum of the mother and only one of the twins contained antibodies to intrinsic factor and gastric parietal cells. Lymphocytes from the mother and the twins, cultured in the presence of intrinsic factor, underwent transformation with increased uptake of H^3-thymidine into DNA (stimulation indices—9.8, 2.5, 1.4). Production of migration inhibition factor in similar cultures in these three patients resulted in 17, 37, and 9% inhibition of macrophage migration. These studies suggest that cell-mediated immunity, rather than humoral immunity, may play a critical role in the development of PA.

Lymphopenia, dysgammaglobulinemia and decreased cellular immunity: A genetic lymphocyte defect. ARMOND S. GOLDMAN, C. WAYNE SMITH, and ELTON DUPREE. *Univ. of Tex. Med. Branch and Shriner's Burns Inst., Galveston, Tex.*

A previously unrecognized genetic immunologic disorder is reported. The patient a 13-year-old female, presented with recurrent viral and bacterial infections, eczema and acute allergic reactions. Humoral and cellular immunity were reduced, the serum IgG and IgM were low; IgD was normal; IgA and IgE were increased. She was lymphopenic, but her thymus appeared normal microscopically. In comparison to normal lymphocytes, her blood lymphocytes in culture did not suppress the life-span of PMNs. In those leukocyte cultures, the PMNs inhibited blastogenesis due to phytohemagglutinin, concanavalin A, pokeweed mitogen and antigens. However, in PMN-depleted cultures the relative frequency of blastogenesis became normal but lymphocyte survival and quantitative blastogenesis due to antigens and mitogens were reduced.

The parents' lymphocytes were unable to limit PMN survival *in vitro* and the relative frequency of transformation of their lymphocytes was reduced. However, the functions of their lymphocytes in PMN-depleted cultures seemed normal. These findings suggest that the principal defect in this disease is in the lymphocytes and that the propositus is homozygous for the defective gene, whereas, the asymptomatic carriers are heterozygous. Although this disorder resembles the Wiskott-Aldrich syndrome, the genetic aspect of the disease, the lack of thrombocytopenia, the low IgG and other features indicate that this is a previously undescribed primary immunologic disorder.

Macrophage function in severe combined immunodeficiency. RICHARD A. GATTI and ROBERT A. GOOD. *Univ. of Minn., Minneapolis, Minn.*

In patients with severe combined immunodeficiency (i.e., lymphopenic agammaglobulinemia), both humoral and cell-mediated forms of immunity are lacking as a result of absence of immunoblasts or lymphoid precursor cells capable of differentiating into plasma cells and small lymphocytes. The small lymphocytes of these infants do not respond to stimulation in vitro with phytohemagglutinin, allogeneic cells or antigens. This situation can be corrected by transplanting bone marrow from an HL-A compatible healthy donor. This finding establishes that the differentiative sites are intact. Macrophage function has not been evaluated in these patients. Nor is there clear evidence to indicate whether macrophages derive from the same defective precursor stem cells as lymphocytes. In these experiments, a glass-adherent cell population was obtained from two patients with severe combined immunodeficiency. After three days incubation, these adherent cells (i.e., macrophages) were added to highly purified lymphocytes from two PPD-sensitive donors. Such highly purified lymphocytes do not respond to antigens in the absence of at