

NEUTROPHIL CELLULAR CHEMOTAXIS IN CHILDREN WITH DOWN'S SYNDROME. James R. Humbert, K. Michael Hambidge, Linda Moore, Benjamin Martinez and Janet M. Stewart. University of Colorado Medical Center, Dept. of Pediatrics, Denver, Colorado.

Neutrophil (PMN) cellular chemotaxis was investigated in 8 children with Down's syndrome, their mothers and 13 adult controls. The new method used assessed separately the number of PMN's adhering to the Millipore filter of a Boyden chamber ("filter chemotaxis", FC) and the number of PMN's migrating into its lower compartment ("compartment chemotaxis", CC). "Total chemotaxis" (TC) was derived by adding FC and CC. Values are reported as percentage of the initial PMN number introduced into the chambers (X ± SEM):

Subjects (#)	FC	CC	TC
Controls (13)	6.5 ± 2.9	20.8 ± 2.7	27.7 ± 2.8
Mothers (8)	8.2 ± 2.3	27.0 ± 5.6	35.2 ± 5.4
Patients (8)	2.6 ± 0.7*	23.4 ± 7.3	25.9 ± 7.4

* P < 0.05 when compared with the other two groups

TC, which assesses best the chemotactic behavior of PMN's, was normal in all subjects, except a single patient who showed complete absence of cellular chemotaxis. Patients had significantly lower FC values than the control groups. Rather than indicating poor chemotaxis, this finding probably reflects decreased PMN adhesiveness to the Millipore filter. Abnormal PMN adhesiveness may contribute to the poor host defense against infections in Down's syndrome.

THE IMMUNOLOGIC ROLE OF TONSILLAR TISSUES IN LOCAL CELL-MEDIATED IMMUNE RESPONSES. RodRigo C. Hurtado, Marek Rola-Pleszczynski, Marco A. Merida, Sally A. Hensen, Monroe M. Vincent, Y.H. Thong and Joseph A. Bellanti. Georgetown Univ. Sch. of Med., Dept. of Ped., Washington, D.C. and Microbiological Associates, Bethesda, Maryland.

The present studies were performed to compare local cell-mediated immune (CMI) responses of tonsillar lymphoid tissues with those of systemic CMI employing lymphoproliferative responses to PHA, T-rossette formation, and specific rubella CMI studies using a ⁵¹Cr lymphocytotoxic microassay. Suspensions of peripheral blood lymphocytes (PBL) and tonsillar tissue lymphocytes (TTL), obtained from 12 subjects ranging in age from 5 to 22 years, were purified by hypaque-ficoll sedimentation and adjusted to equal concentrations. The mean ± SD responses to PHA were 56,642 ± 10,329 cpm for PBL and 38,851 ± 6,804 cpm for TTL (p > 0.6); the mean ± SD values for T-rossette formation were 30 ± 1.7% for PBL and 36.4 ± 3.8% for TTL (p > 0.1); the mean ± SD rubella specific immune release was 13.8 ± 3.7% with PBL compared to 12.6 ± 3.3% for TTL (p > 0.9). No correlation was demonstrated between serum rubella HAI antibody titers and local or systemic rubella specific CMI or between local and systemic rubella specific CMI. These results provide further evidence for the role of local CMI to viruses at mucosal surfaces and suggest the participation of tonsillar tissues in these responses.

VARIABLE PHENOTYPIC EXPRESSION IN A FAMILY WITH "CANDIDA ENDOCRINOPATHY SYNDROME". Sara Kaffe, Linda T. Cahill, and Photini S. Papageorgiou. (Intr. by Philip R. Glade). Mt. Sinai School of Medicine, Dept. of Peds., New York, N.Y. 10029.

The candida endocrinopathy syndrome (CES), an autosomal recessive disorder, has been associated with endocrinopathies and a variety of cell mediated immune defects (CMI). The cell mediated immune responses of a family of 9 members, 3 of which are affected with CES have been investigated by 1) *in vivo* skin reactivity to candida extract (CE), phytohemagglutinin (PHA) and dinitrochlorobenzene (DNCEB); 2) T and B cell rosetting; 3) *in vitro* lymphocyte response to PHA and CE; 4) release of migration inhibition factor (MIF) in response to PHA and CE; 5) the effect of patients' sera on the *in vitro* response of normal lymphocytes to CE. Results are summarized as follows

Pt.	Skin Tests			T Cells	In Vitro PHA		In Vitro CE**		Serum Effect
	CE	PHA	DNCEB		LT*	MIF	LT	MIF	
1	-	+	-	low	+	+	±	-	+
2	+	ND	ND	low	+	+	±	-	+
3	+	ND	ND	low	+	+	-	-	-
other	+	ND	ND	nl.	+	+	+	+	-

*lymphocyte transformation. **cultured in heterologous serum. No abnormal CMI responses were detected in other non-affected members. All the affected individuals had defects in CMI functions. Although it has been reported that family members with CES demonstrate the same defects in CMI, we have demonstrated a variety of defects in this sibship, indicating variable expressivity of the gene involved.

MALIGNANT T LYMPHOBLASTS BIND HUMAN IMMUNOGLOBULIN (Ig), Joseph Kaplan, Jan Cejka, Ward J. Peterson, Jr., Wayne State Univ. Sch. of Med., Children's Hosp. of Mich., Child Res. Ctr. of Mich., Dept. of Ped., Detroit. (Intr. by W.W.Zuelzer)

Human sera were tested by indirect immunofluorescence (IF) for antibodies to cells from 2 malignant T cell lines. By direct IF, these cells, like normal T cells, lacked surface Ig. However, when incubated with normal sera and then treated with fluoresceinated polyvalent anti-Ig, many cells showed patchy surface fluorescence, while similarly treated normal T cells from thymus and peripheral blood were negative. Surface fluorescence was also seen when malignant T cells obtained directly from 2 children with lymphoblastic lymphoma were first incubated in their own sera as well as in control sera. This indicates that positive staining reactions were not necessarily due to alloantibodies. Reactivity seemed related to serum Ig levels since negative or diminished reactions were seen with sera from many newborns and from patients with agammaglobulinemia, while sera from newborns with antenatal infections and elevated Ig levels gave positive reactions. When, following serum treatment, cells were stained for IgG, IgA and IgM, some sera resulted in positive staining for only one Ig class, whereas other yielded positive staining for 2 or all 3 immunoglobulins. These findings suggest that malignant T lymphoblasts differ from normal T cells by possessing surface receptors for Ig. These receptors may represent tumor-related antigens recognized by both the tumor host and by most normals.

EPSTEIN-BARR VIRUS (EBV) NEGATIVE HUMAN MALIGNANT T LYMPHOCYTE CELL LINES. Joseph Kaplan, Thomas C. Shope, Ward D. Peterson, Jr., Wayne State Univ. Sch. of Med., Child Res. Ctr. of Mich., Children's Hosp. of Mich., Dept. of Ped., Detroit. (Intr. by Adnan Dajani)

Most established human lymphocyte cell lines have properties of B cells and are EBV positive. Recently a few lymphoblastoid lines have been shown to have T cell properties (Minowada, J., et al, J.N.C.I. 49:891, 1972) (J. Kaplan, et al, Cancer Res. in press, 1974.) This report describes studies of the malignant lymphoblast cell lines CCRF-CEM and HSB-2, derived from two children with leukemia secondary to lymphosarcoma. Like normal T cells these two lines form more rosettes with sheep erythrocytes at 4° than at 37°. Rosette formation is inhibited to a greater degree by rabbit antiserum to T cell lines than by antiserum to B cell lines. The cells lack complement receptors and surface immunoglobulins. When examined for EBV capsid antigen and EBV nuclear antigen, an indicator of EBV genome, CCRF-CEM and HSB-2 are negative, whereas all B cell lines so studied are positive for EBV nuclear antigen.

These findings indicate that lines CCRF-CEM and HSB-2 are T cells and that they are EBV negative. Perhaps they possess genetic material of some other virus which, like EBV, induces both sustained *in vitro* cell growth and *in vivo* tumors. If so, the availability of these malignant T lymphocyte lines may facilitate the search for a human leukemia-lymphocyte virus.

EFFECT OF ESTRADIOL(E) ON IMMUNE COMPETENCE. Jean F. Kenny and Pamela C. Pangburn, Children's Hosp. of Pittsburgh, Pittsburgh.

Human females have higher serum levels of IGM antibody than males; this sex difference is greatest in late childhood and adulthood. In Swiss mice which also demonstrate sex differences in immunity, females have more splenic antibody producing cells (SAPC) than males, a phenomenon dependent on estrogen secretion. The purpose of this study was to determine when in the antibody producing process E exerts its effect. A single physiologic dose of E (2.5µg) was given to male mice at times from 2 days prior to 4 days after the injection of 5x10⁷ heat-killed *E.coli* (HKE). Mice were sacrificed 4 days after the antigen and counts of anti-*E.coli* SAPC compared to those of littermate controls. Greatest increases in SAPC were observed in groups of 65 mice receiving E 1-3 days after the antigen (p < .01). Significant increases (p < .05) were observed in 42 mice receiving E as late as 12 hrs before sacrifice, but not in those given E 1-2 days before the antigen or 2 hrs before killing. Spleen cells from mice injected with HKE 3 days before were grown for 18 hrs *in vitro*. SAPC or uptakes of H3 thymidine were compared in the same suspensions incubated with and without E (500pg/ml). In 33/44 suspensions treated with E, SAPC were increased a mean of 61%; cpm of H₃ were an average of 40% greater in 10/10 such suspensions. Findings suggest that small amounts of E increase the rates of replication and DNA synthesis of antigen-stimulated spleen cells. These effects which may be observed late in the proliferative phase of antibody production appear to be independent of the enhancing effect of E on phagocytosis.