848 DIFFERENTIAL IN VITRO EFFECT OF SELENIUM (Se) ON NATURAL KILLER (NK) CELL ACTIVITY OF HUMAN LYMPHOCYTES. Madhavan P.N. Nair and Stanley A. Schwartz. The University of Michigan, Depts. of Pediatrics and Epidemiology, Ann Arbor, MI.

Se is an essential micronutrient in humans and animals which plays a key role in many biological processes including the immune response. The in vitro effects of Se as sodium selenite on target binding and lytic activities of lymphocytes were investigated. A human erythroleukemia cell line, K562, and a human histiocytic lymphoma cell line, U937, were used in a 4 hrs ^{51}Cr release assay and in single cell assay in agarose to determine target cell binding and cytotoxic activity. Normal lymphocytes precultured for 48 to 72 hrs with as low as 5 to 200 ng Se/ml produced significant enhancement of their NK activity as well as target binding capacity. Lymphocytes precultured with Se at higher but non-toxic concentrations ranging from 400 to 800 ng/ml showed significant inhibition of NK activity. Inhibition of cytotoxicity by Se was independent of target binding by effector cells, thus indicating that its mode of action is on post-binding functions. These studies suggest that Se has a bimodal immunoregulatory effect on the NK activity of human lymphocytes. These findings may be of significance in developing nutritional protocols for the treatment of primary and secondary immunodeficiency states.

849 EFFECTS OF PYOCYANINE (PYO) AND 1-OH-PHENAZINE (OHP) ON T-LYMPHOCYTE ACTIVATION. J. Nutman, R.L. Waller, M. Berger and R.U. Sorensen, Case Western Reserve U, Dept. of Pediatrics, Cleveland, OH.

Phenazine pigments secreted by *P. aeruginosa* inhibit lymphocyte blastogenic responses. We studied the mechanism of inhibition of two model compound, PYO and OHP on the T-cell cycle. Peripheral blood mononuclear cells (PBMC) were stimulated with the mitogen, Concanavalin A (Con A) or with the following steps of T-cell activation were studied: 1. Increase in cytosolic-free Ca^{2+} using Quin-2 fluorescence. 2. Production of Interleukin 2 (IL-2) using the CTLL-20 cell line. 3. Expression of IL-2 receptors (IL-2R) using monoclonal antibodies and flow cytometry. 4. Uptake of ^{3}H-Thymidine (^{3}HTdR). Results with Con A activated PBMC are shown in the table.

	$\Delta[Ca^{2+}]i$,nM	IL-2,U/ml	IL-2R,%	^{3}HTdR, cpm
Con A alone	207 ± 58	51	56±2	38641±3890
+ PYO (50 μM)	260 ± 105	5	3±1	87±40
+ OHP (100 μM)	102 ± 40	not done	54±3	8600±2853

Lower concentrations of PYO and OHP inhibited Con A-induced ^{3}H TdR uptake synergistically. In ionomycin-activated blastogenesis, ^{3}H TdR uptake was strongly inhibited by PYO (94%), but only to a minor degree by OHP (30%). Therefore, PYO and OHP inhibit T-cell activation at different steps of the signal transduction pathway. OHP blocks the increase in cytosolic Ca^{2+} and PYO acts at a later step not by-passed by ionomycin. Locally secreted phenazine pigments, even in small amount, may act synergistically to inhibit cellular immune responses necessary to eradicate chronic infections with *P. aeruginosa*.

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WITHDRAWN

▲851 MOTILITY OF HUMAN MILK LEUKOCYTES IN COLLAGEN GELS. Fatih Ozkaragoz, Helen E. Rudloff, Srinivasan Rajaraman, Frank C. Schmalstieg, & Armond S. Goldman, University of Texas Medical Branch, Departments of Pediatrics and Pathology, Galveston, TX.

We previously reported that adherence, orientation and directed movement of human milk leukocytes (HMLs) were less than peripheral blood leukocytes (PBLs). Since the diminished motility may have been due to a decrease in adherence, a collagen gel (CG) system where leukocyte movement is less dependent on adherence was used to explore these questions.

Unfractionated HMLs or fractionated PBLs were placed on CGs in microwells and the leading edge of migration was determined by inverted phase microscopy. The mean rates of invasion of HMLs, blood neutrophils and mononuclear blood leukocytes were 20, 200, and <1 μ/H, respectively ($p<0.01$). We then examined the identity of motile HMLs by immunoperoxidase techniques by using antibodies to selected cell markers. Motile HMLs were positive for neutrophil and monocyte markers Mac-1, lysozyme, and a specific macrophage marker cathepsin B, but were negative for markers for lymphocytes (Leu1), NK cells (Leu7) and neutrophils (cathepsin G). The motility of these cells was inhibited (<1μ/H; $p<0.001$) when unfractionated HMLs were stimulated with a T cell lectin (PHA) to produce lymphokines, e.g. migration inhibitory factor. Thus, the diminished motility of milk neutrophils does not appear to be due to decreased adherence per se. Furthermore, those HMLs which are motile are macrophages. The findings suggest a dichotomy for the function of HMLs. Neutrophils may be relegated to the lumen of the alimentary tract, whereas macrophages may penetrate into mucosal sites for host defense.

852 COMPARISONS OF SERUM LEVELS OF KERATAN SULFATE (KS) WITH HEIGHT IN AN OUTPATIENT PEDIATRIC POPULATION (GENPEDS), CONSTITUTIONAL GROWTH DELAY (GD), AND IN JUVENILE RHEUMATOID ARTHRITIS (JRA). Lauren M. Pachman, Jennifer Hayford, Patricia Lynch, Janet Jacobitz, Northwestern Univ. Med. School. Children's Memorial Hospital, Dept. Pediatrics, Chicago, IL; Mary Ellen Lenz, Klaus Kuettner, Eugene Thonar, Rush Presbyterian-St. Luke's Med. School, Dept. Biochemistry, Chicago, IL.

We had previously reported that serum levels of KS increased from birth, reached a plateau between 6-11 years of age and then rapidly declined to levels which were maintained throughout normal adulthood. The purpose of this study was to determine if serum levels of KS were correlated with the percentile height in children in the 6-11 year old age range, using an ELISA with a monoclonal antibody 1/20/5-D-4 to KS. Sera were obtained from 33 GENPEDS whose records had been screened and who had no evidence of autoimmune disease. Levels of KS (ng/ml ± SD) were higher in taller children (≥75th percentile) than shorter children (≤25th percentile): 632±122 vs. 480±126, ($p<.007$). Fourteen children with GD (<5th percentile) had KS values of 414±118 which were not significantly different from the shorter group of GENPEDS, $p=0.11$. In contrast, the GD group had KS values significantly different from the ≥75th percentile GENPEDS, $p=0.001$. KS values in 10 JRA children, ages 6-11, of varying percentile height prior to any steroid treatment were low, i.e. 451±98. Another group of JRA children, on steroids, had an even lower KS value; 399±83. These studies suggest that KS values are correlated not only with the height of the child but may also reflect alterations in cartilage proteoglycan metabolism in JRA which is accentuated by prednisone.

853 IgA RECEPTOR MEDIATED ACTIVATION OF HUMAN MONOCYTES. Shai Padeh, Justen H. Passwell (Spon. by Harvey R. Colten); Sackler School of Medicine, Sheba Medical Centre, Laboratory of Pediatric Immunology, Tel Aviv.

Immune complexes of the IgA isotype are clearly implicated in the immunopathogenesis of several disorders in the pediatric population particularly IgA nephropathy and Henoch-Schonlein disease. Therefore we have studied the interactions of IgA and human monocyte monolayer cultures. Secretory IgA derived from human breast milk and/or IgA derived from an EBV B cell line were used in these studies. Specific binding of IgA to monocyte membrane receptors was confirmed by 1) dose dependent inhibition of IgA-EA rosette formation (control 77±3%; % inhibition IgA 50 μg/ml-88±4, IgA 5 μg/ml-64±15) but not IgG. 2) Iodinated IgA bound to monocyte monolayers. Binding was dependent on the number of monocytes, reached saturation with increasing amounts of ^{125}I-IgA and could be inhibited by unlabelled IgA. Incubation of monocyte monolayers in the presence of increasing concentrations of secretory IgA and $F(ab')_2$ anti-IgA resulted in a dose dependent increase of the oxidative burst (table). Neither IgA or anti-IgA alone nor incubation of IgG with anti-IgA had any effect on the oxidative burst. These studies indicate that human monocytes have a receptor for IgA and that specific activation of the monocytes may occur via these receptors.

Addition	μ mol H_2O_2/mg protein/90 min
IgA 0.1 (ng/ml) +anti-IgA	71±29
IgA 1.0 (ng/ml) +anti-IgA	91±15
IgA 10 (ng/ml) +anti-IgA	159±22
IgA 100 (ng/ml) +anti-IgA	209±22
PMA (20 nM)	182±29