890

ERYTHROCYTE PROTOPORPHYRIN (EP) IN THE NEWBORN-INCREASED RATIO OF FREE TO ZINC COMPLEXED EP IN YOUNG RBC. Ray Yip, Clark M. Smith II, Samuel Schwartz, Betty Stephenson, David P. Tukey, Peter R. Dallman. University of Minnesota, Dept. of Peds and Med., Minneapolis, University of California San Francisco, Dept. of Pediatrics, San Francisco.

EP exists in both the free state (FEP) and in a zinc complexed form (ZEP) in RBC. The ratio of FEP to ZEP in healthy individuals averages 1:10. In the newborn period and in conditions with increased RBC turnover the FEP/ZEP ratio becomes elevated. Using dextran or stractan gradients, cord blood was centrifugally separated into 4 fractions known to contain RBC populations of different ages. The characteristics of these fractions and their EP contents are as follows: (N=6, EP expressed as $\mu\text{g/dl}$ RBC).

#	Retic	MCV	MCHC	Total EP	FEP	FEP/EP	FEP:ZEP
1	19.0%	111	29.3	106	19	17.9%	1:5
2	5.8%	102	33.8	83	7	8.6%	1:11
3	3.9%	96	36.1	91	3	3.2%	1:29
4	4.8%	92	39.4	108	3	2.9%	1:35

The total content of EP did not vary significantly according to RBC age but there was an increased FEP/EP in the fraction of younger, reticulocyte-rich cells. This EP pattern suggests that the newly formed EP is in the free state and may become zinc complexed or lost from RBC with time. The previous observation of an elevation in the free form of EP in hemolytic disorders is likely to be related to the presence of a larger percent of young RBC.

891

ERYTHROCYTE PROTOPORPHYRIN (EP) AS A SCREENING TEST FOR IRON DEFICIENCY IN AN URBAN PEDIATRIC CLINIC.

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EP is the primary screening test for lead poisoning but also becomes elevated with iron deficiency. The effectiveness and limitations of EP in screening an unselected population for iron deficiency has not been fully evaluated. During a 1 yr period 2150 children under 12 yrs were screened with EP during their routine physical. Ten % of the children had EP > 35 ($\mu\text{g/dl}$ whole blood) and were followed up to determine the cause of EP elevation. Iron deficiency was defined as serum ferritin < 15 ng/ml and/or transferrin saturation < 16%. Elevated blood lead was based on two successive blood lead > 30 $\mu\text{g/dl}$. Of 191 children found to have EP > 35 and completing study the findings were:

Iron deficiency only	68.4%
Iron deficiency with elevated blood lead	19.3%
Elevated blood lead only	3.1%
Anemia of chronic disorders	2.1%
Sickle cell anemia	0.5%
Subjects with no abnormal findings (false +)	6.3%

Using EP > 35 as the screening level, 87% of the screen positive subjects had biochemical evidence of iron deficiency. Approximately half of these subjects also had microcytosis or anemia, more strongly indicative of iron deficiency. An EP > 35 was associated with iron deficiency with sufficient frequency to justify further evaluation or a therapeutic trial with iron.

892

HYPOFERRINEMIA IN THE TOXIC-SHOCK SYNDROME (TSS).

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Serum ferrikinetics in the non-bacteremic TSS were examined in 10 female patients (pts) fulfilling the criteria for the Wisconsin case definition. The mean serum iron (Fe) $\pm$ SE levels for the first 5 days of hospitalization are shown (N1=30-180 $\mu\text{g/dl}$). The levels on day 3 (D3) were significantly greater ($P < .001$) than those on D1. Despite a fall in serum total protein (< 5.6

Day of Hospitalization	Mean Serum Fe: $\mu\text{g/dl}$	n	g/dl in 19/20 pts) and albumin (< 3.1 g/dl in 18/20 pts), the total iron binding capacity was within the low normal range (220-480 $\mu\text{g/dl}$) in 5/5 pts. There was no evidence of pre-existing anemia in any of the pts; MCV=88.9 μ^3 $\pm$ 5.0 μ^3 (n=10). (N1 = 77-87 μ^3).
1	10.5 $\pm$ 1.29	10	
2	14.5 $\pm$ 3.81	6	
3	46.3 $\pm$ 10.10	6	
4	111 $\pm$ 20.95	6	
5	91.3 $\pm$ ---	3	

No significant correlation could be made between the Fe levels and the degree of fever or the total number of WBC, PMN or band cells. The rapid increase in Fe paralleled the return of temperature, WBC and band cell values to normal.

Hypoferrinemia, a host response to infection may be due to leukocyte pyrogen induced lactoferrin release from the PMN series cells and increased RES storage of Fe (Weinberg ED, Microbiol Rev 42:45, 1978). The high temperatures and marked increase in cells of the PMN series (90.6 $\pm$ 8.8%; n=10) in TSS may be responsible for the profound hypoferrinemia, which may provide a marker for disease activity.

IMMUNOLOGY

● 893

MORPHOLOGY, DEVELOPMENT AND DISTRIBUTION OF HUMAN NK CELLS IDENTIFIED BY A MONOCLONAL ANTIBODY (HNK-1). Toru Abo, Max D. Cooper and Charles M. Balch. Cellular Immunobiology Unit, University of Alabama in Birmingham

A monoclonal IgM antibody (HNK-1) has been shown to react with a subset of human lymphocytes that can function as natural killer (NK) and antibody dependent killer (K) cells. HNK-1 antigen is present on 60% of T γ cells (Fc γ receptor $^+$, E-rosette $^+$) and 56% of "null" cells (Fc γ R $^+$ ER $^+$ sig $^-$) but not on sig-bearing B cells and monocytes. In the present studies we have used immunofluorescent and cell sorter techniques to examine the development, distribution, morphology and function of cells expressing HNK-1. Fewer than 1% of cells in thymus and newborn blood samples were HNK-1 $^+$. The number of circulating HNK-1 $^+$ cells increased as a function of age: <5% of blood mononuclear cells in children under 15, 5-35% in adults 15 to 40 and 25-45% in adults >40 years of age. HNK-1 $^+$ cells comprised a mean of 12% of spleen cells but <1% of lymph node cells. The level of NK and K cell functions was closely related with the numbers of HNK-1 $^+$ cells in all tissues examined. Morphologically, HNK-1 $^+$ cells were defined as a homogeneous population of lymphocytes with abundant cytoplasm containing azurophilic granules. A significant proportion of HNK-1 $^+$ cells coexpressed antigens previously defined by monoclonal antibodies on T cells and monocytes. Our studies define a morphologically distinct subpopulation of lymphocytes of unknown origin with NK and K activities, the HNK-1 $^+$ cells were shown to expand as a function of age and exhibit selective migration. NIH CA 16673 and CA 27197.

● 894

IMPAIRED CHEMOTACTIC FACOTR (CF)-INDUCED REDISTRIBUTION OF ADHESION SITES IN NEONATAL (N) PMNs. D.C. Anderson & C.W. Smith. Baylor College of Medicine, Houston, TX 77030, & Mich. State Univ., E. Lansing, MI 48824.

CF sensory and effector PMN functions were studied in 75 healthy N & adult controls (C). N PMNs demonstrated saturable high affinity binding for f-Met-Leu-Phe (fMLP)=C. One-half maximal binding by N & C PMNs occurred with 12 nM fMLP @ 37°C; # binding sites/PMN=4050 $\pm$ 900(N), 3650 $\pm$ 750(C). Maximal CF stimuli (5X10 $^{-8}$ M fMLP) induced uropod formation in only 38 $\pm$ 8% of N PMNs compared to 73 $\pm$ 11% of C PMNs ($p < .01$). Several abnormalities of N PMN adherence (Ad) were identified; diminished Ad increments above baseline were noted in response to single CF stimuli (fMLP; 42 $\pm$ 78(N), 70 $\pm$ 11(C), C5a; 41 $\pm$ 6(N), 68 $\pm$ 6(C); $p < .01$. On sequential CF exposure, N PMNs failed to demonstrate decreased Ad values: fMLP (10 $^{-10}$ M, 10 $^{-8}$ M); 51 $\pm$ 9(N), 30 $\pm$ 9(C), C5a (8 $\mu\text{g/ml}$, 32 $\mu\text{g/ml}$); 48 $\pm$ 14(N), 34 $\pm$ 10(C); $p < .001$. As shown by latex bead binding, N PMNs also failed to redistribute adhesion sites to cell uropods with sequential CF stimuli; restricted uropod binding occurred in 33 $\pm$ 8% (fMLP) or 37 $\pm$ 7% (C5a) of N PMNs in contrast to 70 $\pm$ 6% (fMLP) or 71 $\pm$ 8% (C5a) for C; $p < .001$. Similar findings were shown with colchicine (25 $\mu\text{g/ml}$) incubation; diminished Ad (relative to baseline) was shown with C (-17 $\pm$ 6%) but not N PMNs (-4 $\pm$ 6%) ($p < .001$), and uropod binding by N PMNs (31 $\pm$ 11%) was significantly ($p < .001$) less than C (72 $\pm$ 14%). Conclusion: CF sensory mechanisms of N PMNs are normal. Diminished stimulated locomotion of N PMNs is functionally related to impaired redistribution of their adhesion sites by CFs.

● 895

ANTIBODY-INDEPENDENT CLASSICAL PATHWAY (CP)-MEDIATED OPSONOPHAGOCYTOSIS (OP) OF TYPE Ia GROUP B STREPTOCOCCI (IaGBS). Carol J. Baker, Morven S. Edwards, Bette J. Webb and Dennis L. Kasper. Baylor College of Medicine, Houston, and Channing Laboratory, Harvard Medical School, Boston.

Since invasive IaGBS disease rarely occurs beyond the early neonatal period, heat-labile serum factors rather than type-specific antibody (SpAb) could be critical to host defense against IaGBS. The OP requirements for prototype strain, 090, and 18 fresh clinical isolates were assessed in an assay employing human PMN, serum containing endogenous complement, and 3x10 6 CFU/ml of IaGBS. Significant OP of 090 (>90% reduction in CFU) occurred after 40 min. incubation in 6 sera containing high levels of SpAb, while bacterial growth was noted in 25 sera with low levels. The mean OP for 090 was compared to that for 18 clinical isolates in 3 representative sera. OP was similar in serum with high SpAb levels (94% vs 97%). However, in low SpAb and agammaglobulinemic sera, respectively, OP was 94% (range, 83-98%) and 95% (range, 77-99%) for 18 clinical isolates, while 090 grew in these sera. Absorption of serum with high SpAb with Ia polysaccharide antigen (Ag) completely inhibited OP for 090, but did not effect that for clinical isolates. All strains required high SpAb levels for alternative pathway recruitment. Serum deficient in C2 and SpAb did not promote OP of clinical isolates of IaGBS until reconstituted with C2. These findings that OP of clinical isolates of IaGBS is CP-mediated in a non-SpAb dependent fashion indicate that substances other than Ia capsular Ag, such as polar glycopospholipids, may initiate CP activation in the absence of immunoglobulin G.