

EFFECT OF PARACHLOROMERCURIBENZOATE (PCMB) ON PLATELET FUNCTION AND METABOLISM AT pH 5.3.

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The effect of different SH blockers on blood platelets at low pH has been studied. Washed human platelets secrete 70% of preabsorbed serotonin at pH 5.3 when exposed to 1 mM NaF for 5 min at 37°C. The secretion is 60% inhibited by 0.1 mM PCMB. With higher concentrations of PCMB the extracellular concentration of preabsorbed serotonin rises to 75%. In the absence of fluoride 0.1 mM PCMB causes liberation of 43% of preabsorbed serotonin, while the extracellular [serotonin] found with higher [PCMB] alone does not vary from the values found in the presence of NaF and PCMB. At the same time cytoplasmic nucleotides are found extracellularly, but amount to only 1/2 to 1/3 of the liberated serotonin. This indicates that PCMB inhibits secretion induced by fluoride, while at the same time causing selective liberation of stored material from platelets by a process which may or may not be a secretory process. In contrast to NaF, 3 min exposure to 0.2 mM PCMB produces no distinct ultrastructural changes and no acceleration of the effect of exposure to pH 5.3. In the presence of PCMB the breakdown of metabolic ATP is impaired, and there is no production of inosine and hypoxanthine. PCMB (0.1-0.3 mM) does not induce any of the described changes when added at pH 7.4. When, however, the platelets are first incubated with PCMB at pH 5.3, then the changes induced by the compound continue at pH 7.4. Neither N-ethylmaleimide nor parachloromercuriphenylsulfonic acid induce similar changes at the low pH. The platelet membrane seems therefore to be specifically receptive to the effect of PCMB at this pH, or PCMB itself has been converted to a form to which the platelet membrane is sensitive.

THE SOURCE OF HYDROGEN PEROXIDE AND OF CHEMILUMINESCENCE OBSERVED IN ACTIVATED HUMAN PLATELET PREPARATIONS. P.H. Levine, K.L. Scoon, J.C. Hardin and N.I. Krinsky. The Memorial Hospital and the University of Massachusetts Medical School, Worcester, and Tufts University School of Medicine, Boston, Massachusetts, U.S.A.

Human platelet suspensions can be observed to produce small amounts of H_2O_2 (.04 nmoles H_2O_2 /min/ 2.5×10^5 cells mm^{-3}) and measurable chemiluminescence when exposed to target particles for phagocytosis, such as latex spherules. Both H_2O_2 production and chemiluminescence are characteristic of phagocytosing polymorphonuclear leukocytes (PMN) and analysis of the purified platelets indicates contamination by PMN at the level of 0.2%. The amount of H_2O_2 produced and the chemiluminescence observed can be duplicated by adding latex spherules to a preparation of PMN at a concentration equivalent to the contaminant in the platelet preparations. We conclude that the H_2O_2 produced and chemiluminescence observed from activated platelets is due to the presence of small amounts of contaminating PMN. The production of H_2O_2 and free radicals by the PMN which contaminate platelets may contribute to loss of platelet function during storage. These studies also indicate the importance of controlling for PMN contamination in studies of platelet biochemistry and physiology.

STUDIES ON ANTI-PLATELET ANTIBODIES. ITS CORRELATION WITH ACID PHOSPHATASES. PRELIMINARY RESULTS. H.N. Hendler and E.S. Sack. Fundación Viviana Luckhaus, Hospital Juan A. Fernández, Buenos Aires, Argentina.

The platelet factor 3 immuno-injury technic (Karparkin and Siskind, Blood 33:795, 1969) has been evaluated as a test for the detection of anti-platelet antibodies in 126 patients with quantitative and qualitative platelet disorders. Plasma acid phosphatases in platelet-poor plasma (PAP-PPP) were simultaneously determined in a selected group of patients. Also the effect of serum incubation (from patients and controls) on acid phosphatase availability of platelet-rich plasma (APA-PRP) was investigated.

Anti-platelet antibodies were present in 52 % of patients fulfilling the diagnostic criteria for idiopathic thrombocytopenic purpura and in 38 % of patients with miscellaneous conditions. The following table expresses the results of acid phosphatase determinations in the selected group of patients studied:

Anti-Platelet Antibodies	Acid Phosphatases				Thrombocytopenic
	PAP-PPP		APA-PRP		
	Normal	Elevated	Normal	Elevated	
Negative	0	100	86	14	100
Positive	58	42	42	58	75

Platelet destruction is the probable cause of the thrombocytopenia in the group with negative anti-platelet antibodies. Further, their serum lacks membrane-injury action. On the other hand the results in the group with positive antibodies suggest that heterogeneous immunologic mechanisms are operative.