Authors’ Summaries of Articles Accepted January, 1966


The rate of appearance of C\textsuperscript{14} in the heme and globin of peripheral blood following intravenous administration of glycine-2-C\textsuperscript{14} has been measured in normals, patients with androgen-induced erythrocytosis, polycythemia vera, (iron deficiency), and virilizing Cushing's disease with polycythemia. In all of these cases of accelerated erythropoiesis a common alteration of the globin specific activity curve was observed. Heme specific activity curves were not affected. The changes in the globin specific activity curves are best explained by consideration of the marrow erythroid cellularity during erythrocytosis.


The characteristics of proliferation of leukemic cells in four children with untreated acute leukemia have been studied. In all four of these children a population of marrow leukemic cells was found which were dividing with a generation time of about 15 to 20 hours. In two of these patients it was possible to demonstrate that these dividing cells after one or more mitotic divisions became smaller and stopped dividing. In all of these patients 70 per cent or more of the leukemic cells of the marrow and almost all leukemic cells of the blood were nonproliferative at the time of these studies. These nondividing cells would be relatively unaffected by chemotherapeutic agents designed to inhibit cell division.


Monocytes, defined as peroxidase positive mononuclear cells in the peripheral blood of healthy rats, were labeled by frequent intermittent injections of tritiated thymidine. About 25 per cent of the monocytes were labeled within 1 day and 82 per cent in 8 days. Both labeled and unlabeled monocytes disappeared from the circulation in accordance with an exponential function with a half-time of about 3 days. Mean grain counts increased asymptotically toward a limit reached in 4 or 5 days. The monocyte turnover rate in the rat is in the neighbourhood of 3.6 \times 10^6 cells per day.

It is concluded that monocytes leave the circulation at random and not as a consequence of senescence. It is probable that they are the product of a cell lineage consisting of about 3 generations from the primitive precursor to the mature form and that the average generation time is about 24 hours. Because of the rapid appearance of large numbers of labeled cells, it is unlikely that they are derived from lymphocytes which acquire label much more slowly.


The interaction of human blood platelets with influenza virus (PR-8) was studied in vitro and in vivo.
It was found that “live” influenza virus was rapidly adsorbed onto human blood platelets at 4 C. and completely eluted at 37. “Dead” virus was adsorbed at 4 C. but not eluted at 37 C. unless the platelets were treated with RDE (receptor destroying enzyme). Adsorption of virus also occurred at temperatures above 4 C. (from 20 to 37 C.) However, while adsorption was maintained throughout incubation at 4 C., slow elution occurred after 30 to 90 minutes incubation at 26 to 37 C. Storage of the platelets for lengthy intervals at 4 C. or coating of the platelets with macromolecules did not interfere with virus adsorption. After one cycle of adsorption-elution, blood platelets could not adsorb virus again. Treatment with RDE greatly reduced virus adsorption. During the process of virus adsorption, prominent platelet clumping occurred. During elution, clumping remained unchanged and gross alterations in morphology of the platelets were observed. In the process of virus adsorption-elution, large numbers of platelets were lysed.

Comparative experiments were performed simultaneously with human red blood cells (RBC) and identical results were obtained as with blood platelets. However, the extent of adsorption of live virus was equal for platelets and RBC only when the relationship between platelet number and RBC number in the preparations used was 6:1. This suggested a direct proportion between the surface area of both platelet and RBC and the number of available virus receptors.

Virus suspensions infused into rabbits produced a sharp and sustained drop of the platelet count. Survival of radioactively labeled platelets treated with virus prior to infusion was markedly shortened with live virus and was only slightly reduced with dead virus.

It is suggested from these experiments that blood platelets, as other blood cells, may serve as carriers of viruses in the circulation and that in this process the platelets are damaged and partially destroyed.


The five rabbit anti-rhesus monkey erythrocyte specificities of Owen and Anderson1 have been confirmed. An additional specificity, designated F, has been found in 91.2 per cent of rhesus animals tested. Antiserum reagents with the A and B reactivities have been used in the indirect fluorescent antibody technique to detect and quantitate minor rhesus erythrocyte populations in admixture with a major population. Specific fluorescence was achieved with wet, unfixed preparations and was all-or-none. A rapid method for the estimation of minor to major cell population ratios well above 1:1000 is presented.


1. Six patients in whom Hodgkin’s disease or lymphosarcoma developed during the use of anticonvulsant agents (Dilantin, phenobarbital, Mysoline, Celontin) are reported.
The differential diagnosis from pseudolymphoma, the course of these malignant lymphomas, and their therapeutic management are discussed.

Based upon information presently available, it could not be determined whether the atypical hyperplasia sometimes observed during anticonvulsant drug therapy is precancerous, or whether these agents may be carcinogenic in certain sensitive individuals.

Patients with a seizure disorder who develop lymphoma require standard therapy for both conditions. The anticonvulsant drugs had no evident adverse effect on the course of the lymphoma, although substitution of another agent seems warranted, if possible, for the offending drug.


Although the concentration of erythropoietin may rise to very high and easily measured levels under abnormal circumstances, the concentration in normal animals of many species, including man, is not sufficient to be consistently demonstrated in unconcentrated samples of serum or urine. The fact that the administration to normal animals of antibodies against erythropoietin results in cessation of erythropoiesis is evidence that the hormone is essential for red cell production, and therefore must be present in low concentration in the body fluids of normal animals.

Recent work in this laboratory has shown that a standard method of concentrating urinary erythropoietin and a standard assay procedure can be used to demonstrate the hormone in the urine of normal human beings. The erythropoietically active material recovered produced increasing response to increasing dose. It was completely neutralized by rabbit serum containing antibodies to human urinary erythropoietin. The average normal man excretes approximately 1 standard A unit of erythropoietin per day and the average normal woman excretes approximately 0.4 units per day. With the exception of one patient with a renal allograft and normal subjects living at 17,000 feet, the recovery of erythropoietin from the urine has not been found to exceed normal in patients with polycythemia, whether the polycythemia is primary or secondary to renal pathology. A 20-fold increase in excretion of erythropoietin was found in permanent residents at high altitude (Chacaltaya, Bolivia, 17,000 feet).


The platelets of a patient with congenital thrombasthenia were not aggregated by ADP, thrombin, connective tissue particles, Polybrene, or phospholipase C, and did not adhere to glass as measured either on a glass slide or by retention in a glass bead column. Clot retraction was markedly diminished. Raising the magnesium level partially corrected clot retraction but did not restore ADP-induced clumping. The platelets were less able to promote prothrombin consumption. Fibrinogen concentration in the supernatant of frozen and thawed platelets was low, but surface fibrinogen appeared to be normal.
The thrombasthenic platelets were normal in the following respects: concentration of ATP and glyceraldehyde-3-phosphate-dehydrogenase; adhesion to connective tissue fibers; aggregation by antiplatelet serum; microelectrophoretic mobility; isoelectric point; disc shape of platelets at 37 C.) ability of platelets to change shape with ADP or cold; decrease in ATP concentration and auramine staining of granules by thrombin; release of serotonin, and ADP and other materials absorbing at 260 mµ by thrombin or connective tissue particles; liberation of acid phosphatase during blood clotting; and platelet Factor V activity.

It is concluded that responses of thrombasthenic platelets to thrombin and connective tissue particles are normal except that the liberated ADP fails to cause aggregation. The first stage of the reaction to ADP, transformation from disc to spiny sphere, is normal. Still to be determined at the molecular level are the cause(s) of failure of clot retraction and ADP-induced aggregation and the relationship of these defects to the low fibrinogen concentration of platelet extracts.

Pearson, H. A.: The binding of Cr³⁺ to hemoglobin. II. In vivo elution rates of Cr³⁺ from CC, Hb CS and placental red cells. First submitted Dec. 1, 1965; accepted for publication Jan. 16, 1966.

In vivo elution rates of Cr³⁺ from red cells containing Hb C and from placental red cells containing large amounts of Hb F have been determined. These were found to be 1.8 and 0.85 per cent per day respectively, which do not differ greatly from that of normal adult red cells. Therefore, no special correction factors for elution need be used in evaluating Cr³⁺ survival curves in these clinical situations. The in vivo elution rate of 3.5 per cent per day observed for Hb C-S is significantly greater than that of normal red cells.


1. Three siblings with Hb H thalassemia have been observed in a family in which 17 members from three generations have been examined. Four additional family members have α-thalassemia minor.

2. A genetic examination of the family provides further evidence for the theory that Hb H thalassemia is due to the interaction of the α-thalassemia gene and another "silent" gene which, however, does appear to produce minor morphologic abnormalities when present alone.

3. Reevaluation of the effects of splenectomy indicate that this procedure may be a significant benefit only in the more severe cases of Hb H disease.


Deer mice with a compensated hemolytic syndrome caused by hereditary spherocytosis were placed in a controlled temperature room at 35 C. Severe anemia, red cell fragmentation, increased osmotic fragility, occasional jaundice and hemoglobinuria were observed. The spleen enlarged and the erythropoie-
tic bone marrow expanded. Reticulocytes fell markedly early during heat exposure, but in survivors they soon increased to levels higher than those observed prior to experiment. Mortality from the severe acceleration of the pre-existing hemolysis was high. Control nonspherocytic deer mice subjected to an identical temperature showed only a mild drop of hemoglobin and slight elevation of reticulocytes.

Since hereditary spherocytosis in deer mice in a temperate climate is compatible with good health, these findings provide a further illustration of interaction of a particular environmental insult with a specific genotype in the pathogenesis of hereditary disease.

Beutler, E.: A series of new screening procedures for pyruvate kinase deficiency, glucose-6-phosphate dehydrogenase deficiency and glutathione reductase deficiency. First submitted Nov. 4, 1965; accepted for publication Jan. 10, 1966.

A new type of screening procedure for the detection of enzymatic defects of the red cell has been described. The blood or red cell sample is added to the reaction mixture. After a suitable period of incubation a drop of the mixture is spotted on filter paper, permitted to dry, and examined for fluorescence under UV light. In this way the oxidation of reduction of pyridine nucleotides is readily evaluated. Reaction mixtures for the detection of glucose-6-phosphate dehydrogenase deficiency, pyruvate kinase deficiency, and glutathione reductase deficiency are described. The same general procedure should be readily adaptable to the detection of other enzymatic deficiencies of red cells, such as phosphogluconate dehydrogenase deficiency or triosephosphate isomerase deficiency.


Phagocytosis has profound effects on several aspects of the RNA metabolism of human leukocytes. The major changes induced by particle ingestion appear to be (1) an increased uptake of pyrimidine precursors from the suspending medium, (2) a contraction in the size of the nucleotide pool, (3) an accelerated rate of destruction of preexisting RNA, and (4) an increased rate of RNA synthesis. Sucrose density gradient analysis of the newly synthesized RNA suggests that several classes of RNA are involved in this process. The increased turnover rate of the nucleotide pool and of the cellular RNA of the leukocyte is proportional, within limits, to the total load of ingested particles.


Levels of erythropoietin have been consistently demonstrated in urine collections from healthy adult males using a modification of the polycytemic mouse assay. This method is stable, reproducible and provides assay animals with low
background erythropoiesis. Six-hour collections of urine can be concentrated without apparent loss of activity and the entire daily excretion can be used in the assay. Quantitation has been achieved by comparing the response to a concentrate of normal urine directly to a linear dose/response curve of erythropoietin standard B. Based on 6-hour urine collections, erythropoietin excretion in the subjects tested averaged 4.0 units of erythropoietin standard B (range 1.2–9.5). In addition to wide unexplained variations in results seen from day to day, the data indicate that a diurnal pattern of excretion of the hormone may exist.