ABSTRACTS

Theodore H. Spaet, M.D., Editor

John L. Bateman, M.D., Upton, L. I., N. Y.
Ernest Beutler, M.D., Duarte, Calif.
Jerzy Jozef Biezonski, M.R.C.P.I., New York City
T. H. Bothwell, M.D., Johannesburg, South Africa
T. E. Brittingham, M.D., St. Louis
J. B. Chatterjea, M.D., Calcutta, India
Amos Chernoff, M.D., Knoxville, Tenn.
G. C. deGruchy, M.D., Melbourne, Australia
Pietro deNicola, M.D., Pavia, Italy
Ludvik Donner, M.D., Prague, Czechoslovakia
A. J. Erslav, M.D., Philadelphia
H. Hugh Fudenberg, M.D., San Francisco, Calif.
Katsuhiko Fukutake, M.D., Tokyo, Japan
Robert Goldstein, M.D., Boston
J. Guasch, M.D., Barcelona, Spain
Roger M. Hardisty, M.D., London, England
Victor Herbert, M.D., Boston
Marjorie B. Zucker, Ph. D., New York City, N. Y.

ABSTRACTS OF SPECIAL INTEREST


This paper compares the results of exchange transfusion by the orthodox technic with the results of treatment with 25 Cm. of salt-poor human albumin in the first half of the exchange transfusion in erythroblastosis fetalis. Quantitative estimation of the bilirubin removed was made in each series. A statistically significant increase in bilirubin removal was noted in the albumin-treated group. The possible modes of action of albumin are discussed together with the indications and contra-indications for its use. It is concluded that exchange transfusion with albumin-enriched blood can be expected to reduce the necessity for reexchange, and to increase bilirubin removal at each individual exchange transfusion.—G. C. de G.


The combination of an antiglobulin with the corresponding globulin often manifests itself in a visible manner, for instance by an agglutination, as in the Coombs test. In other circumstances, the combination does not give rise to anything directly detectable, and the reaction has to be "revealed" by measuring the quantity of antiglobulin which has disappeared from the supernatant when the reaction is completed. The antiglobulin consumption test has become quite useful in immunohematology. It enables one to visualize the presence of antiplatelet substances in certain thrombocytopenic purpuras, and of antiplatelet and antileukocyte substances in LE and a few granulocytopenias. It is also useful in tissue immunology, the antigen being constituted by various human tissues which have been homogenized and lyophilized. In rheumatoid arthritis, for instance, the existence of a substance reacting with connective tissue and different from the rheumatoid factor has been demonstrated. In rheumatic fever with heart involvement, the reaction may be positive if heart tissue is employed, and the same in multiple sclerosis with brain tissue. The uses of the antiglobulin consumption test can still be broadened. Until recently, only human anti-D y-globulin was utilized to reveal the consumption of the antiglobulin. But other proteins can be fixed on tanned erythrocytes, and this enables one to demonstrate the consumption of other antibodies reacting with the antiglobulin. The anti-kidney antibody of Masugi's
experimental nephritis is a good example. Also it appears that the antiglobulin consumption test could be employed for the study of delayed hypersensitivity. Experiments have shown that antiglobulin can serve to detect not only the fixation of the antibody on the cell, but also that of the antigen on a cell already coated with the antibodies which are responsible for delayed hypersensitivity.—C. M.


The presence of serum antibody to intrinsic factor (I.F.) was determined by estimating the urinary excretion of Co153-B12 when this was given by mouth alone, with hog pyloric mucosa, and with hog pyloric mucosa plus serum from either normal subjects or from patients with pernicious anemia (P.A.). Antibody could not be demonstrated in 30 normal subjects, but was present in about half of 53 patients with P.A. who had been treated with hog I.F. or a crude stomach preparation by mouth, and in just over a quarter of 33 previously untreated patients. Five patients treated exclusively by injection did not have I.F. antibody. I.F. antibody was shown to be in the globulin fraction and was not species-specific; thus antihog I.F. inhibited rat and human I.F. as well as hog I.F. To evaluate its mode of action further, I.F. antibody was conjugated with fluorescein and tested against human stomach as the antigen, but the results were inconclusive; a tanned-red-cell technique is at present under test. The origin and action of the antibody are discussed, and it is concluded that P.A. may in some cases be an autoimmune disease, that treatment may raise a low pretreatment antibody titer to measurable values, and that the antibody may prevent the binding of the complex B12-I.F. with the acceptor mechanism in the small intestine.—R. M. H.


As a result of starch-gel electrophoresis on 70 cord-blood samples from Negro babies, 3 bloods were found with HbA, HbF, and a fast-moving component I; one with HbA, HbF, and a fast-moving component II; and one with HbA, HbF, HbS, and a fast-moving component II. Subsequent investigation showed that component I (called "Augusta I") was composed entirely of the β chains of S hemoglobin (β4S) and that component II was identical to hemoglobin Barts (γ4F). Family studies in one patient with Augusta I hemoglobin showed that small quantities of hemoglobin H (β4H) were present in the propositus, his father, and four other members of the family who were heterozygous for hemoglobin S, but were not found in two other members who were also heterozygous for hemoglobin S. This family thus appears to have a genetically determined α-chain deficiency which is inherited independently of the β-chain abnormality of the sickle-cell trait. In one family with Bart's hemoglobin the hemoglobins of both parents and seven children were completely normal, and in a second family the father had the sickle-cell trait as the only abnormality. It thus appears that the α-chain deficiency of the first family was demonstrable during both fetal and adult life, but that in the other two families there was an α-chain deficiency affecting only fetal hemoglobin. These findings suggest that the adult α-chain was suppressed in the first family, and the fetal α-chain in the other two; they thus provide evidence that the production of adult and fetal α-chains is controlled independently.—R. M. H.

LEUKOCYTES


It is postulated that the hydroperoxide catalyst complex possesses a sufficient oxidation potential to oxidize benzidine, whereas the effective oxidation potential of O2 catalyst complex has only enough to oxidize leuco indophenol blue. Reduction of hydrogen peroxide in the benzidine reaction and in the non-alkaline naphthol reaction is not due to the donor specificity of the catalyst, but results from sufficient effective oxidation potential to oxidize hardly oxidizable benzidine, or hardly oxidizable naphthol in non-alkaline media. High effective potential of both O2 catalyst and of H2O2 catalyst complex of leukocytes suggests its possible participation in the respiratory chain in leukocytes at the level of cytochrome c oxidase.—K. F.

THE INFLAMMATORY "LEUKOPENIC FACTOR" (MENKIN) AND PHAGOCYTOSIS BY LEUKOCYTES. G. Ludány, Gy. Vajda, A. Döklen and I. Fehér. From the II. Surgical Clinic of the Medical Uni-

Menkin’s inflammatory leukopenic factor, administered intravenously to dogs, inhibits phagocytosis of bacteria by polymorphonuclear granulocytes. In the concentration range of 10^{-4} to 10^{-6} the leukopenic factor also inhibits in vitro phagocytic activity of granulocytes.—S. R. H.


The patient is a 4 year old boy. The blood picture showed 0.5 per cent of neutrophils with monocytosis and lymphocytosis. The periodic alternation of neutropenia was found since one year of age. These regular episodes lasted for three to five days and the interval was about 14 days. During intervals, the patient was without fever and other clinical symptoms without improvement of the blood picture followed administration of prednisolone.—K. F.


Report of three cases of multiple myeloma in persons who had been exposed to x-ray for 25–40 years. Since it is established that leukemia follows x-ray exposure, it is the opinion of the author that all agents which can induce leukemia might also cause myeloma.—H. M.


Serum and urine samples from 25 cases of cytologically confirmed myeloma were donated from several hospitals in Tokyo, and records of 140 cases were given for statistical investigation by hospitals throughout Japan. In the biochemical investigations, the amino acid composition of two cases of M type were analyzed. Although no abnormal amino acids were found, slight differences in composition were seen between myeloma protein and normal γ-globulins. Glycine, glutamic acid and arginine were increased; alanine, tyrosine and phenylalanine were decreased slightly. Optical rotation was tested in H₂O and 2-chloro-ethanol. The myeloma protein folded to 39 per cent as α-helix in 2-chloro ethanol, whereas normal γ-globulin folded up 50–60 per cent as α-helix. In the infrared spectral determination in D₂O, β-forms were observed neither in normal γ-globulin nor in myeloma protein, and β-forms were scarcely differentiated from α-helix by the CO hand. The unfolding, exchangeable N-D₂ groups were 71 per cent in myeloma protein, in contrast to 56 per cent in normal γ-globulin.—K. F.


This is a summary of the results obtained from the treatment of 240 patients with cytostatic agents for malignant disorders of the hemopoietic system. The effects of the following 12 compounds were compared: nitrogen mustard, Degranol, Mitomycin, TEM, Endoxan, Sarcosyl, Sanamycin, Myleran, urethan, colcemid, ACTH, stilbamidine. Degranol produced the most satisfactory remissions in chronic lymphocytic leukemia and lymphosarcoma, and Myleran in chronic granulocytic leukemia. Against reticulosarcoma the cytostatics tested had nearly the same (relatively poor) effect. In multiple myeloma remissions were of short duration. While already equivalent to radiation therapy in a few syndromes, cytostatic agents in themselves are not yet as palliative as is irradiation in others.—S. R. H.

ERYTHROCYTES


Vitamin B₁₂ and the 5,6-dimethyl benzimidazolylcobamide coenzyme were equally active in (1) supporting growth of chicks up to four weeks of age and in (2) overcoming the impaired metabolism of formiminoglutamic acid that occurs in Vitamin B₁₂-deficient chicks.—V. H.

STABILITY OF INJECTED VITAMIN B₁₂-Co60 AND VITAMIN B₁₂ CONTENT OF DOG LIVER. C. Rosenblum, D. A. Willigan, H. T. Meriwether and E.
ABSTRACTS


By reverse isotope dilution, it was demonstrated that injected Co\textsuperscript{60} cyanocobalamin is retained by dog liver as intact cobalamin, probably still in cyano form.—V. H.


Beginning 48 hours after injection, two disappearance curves were found in female rabbits but only one in chickens, male rabbits, and male humans. Comparable tissues from different animals have different B12-binding affinities.—V. H.


Electrophoresis of gastric juice revealed the slowly migrating B\textsubscript{12}-binder to be absent in pernicious anemia. This may be related to intrinsic factor and of diagnostic value.—V. H.

CLEARANCE OF INTRAVENOUSLY INJECTED RADIOACTIVE COBALT-LABELED VITAMIN B\textsubscript{12} IN CHRONIC MYELOID LEUKEMIA AND OTHER CONDITIONS. N. D. Ritz and L. M. Meyer. From Maimonides Hospital, Brooklyn, N. Y. Cancer 13:1000, 1960.

Loading doses of nonradioactive vitamin B\textsubscript{12} converted the delayed clearance to normal, presumably due to saturation of the plasma receptors by nonradioactive B\textsubscript{12} which allowed the radioactive B\textsubscript{12} to go directly to tissue binding sites. It is difficult to interpret most of the experiments because the time intervals separating nonradioactive and radioactive B\textsubscript{12} administration are not stated for each separate experiment.—V. H.


The authors followed the rise and fall of plasma radioactivity after orally administered radioactive vitamin B\textsubscript{12}. They conclude that the most likely mechanism for the increased serum B\textsubscript{12} level in patients with chronic myelogenous leukemia (CML) is a relative shift of B\textsubscript{12} from tissue sites to plasma. This is in accord with the possibility that the high levels of both serum B\textsubscript{12} and serum B\textsubscript{12}-binding protein in patients with CML may be due to release of these materials into the blood stream by leukemic cells with a shortened life-span. The authors mention the high unsaturated B\textsubscript{12}-binding capacity of CML serum as only "an additional factor" in producing the high serum B\textsubscript{12} level. It is possible they give too short shrift to this factor. Their statement that the delayed plasma disappearance of labeled B\textsubscript{12} in CML may be merely an isotope dilution effect seems contrary to the demonstration (Blood 15:646, 1960) that radioactive B\textsubscript{12} mixed with CML serum and injected into a normal recipient has a slow disappearance rate.—V. H.


The effect of chlorpromazine on the absorption of Fe\textsuperscript{55}Cl\textsubscript{3} was studied. Iron absorption from the intestine is depressed by a single dose of chlorpromazine. This inhibition is more pronounced following administration of the drug for 20 hours. Authors assume that dominantly central, and partly peripheral, inhibition of adrenergic impulses may influence iron metabolism, especially iron absorption.—S. R. H.


The Rh\textsubscript{D} (D) antigenic content of the erythrocytes of 199 Caucasians was estimated from the uptake of I\textsuperscript{131} labeled antibody obtained by elution. For Rh\textsubscript{D} (D) positive but rh\textsuperscript{C} (C) negative erythrocytes the amount of bound antibody revealed a bimodal distribution with the lower uptake peak corresponding to the values found for known heterozygotes; the higher peak represented 1.6 times this value and was presumed to correspond with the homozygous expression of Rh\textsubscript{D} (D). For erythrocytes possessing both Rh\textsubscript{D} (D) and rh\textsuperscript{C} (C) only 70–73 per cent of this uptake was noted, and the bimodal distribution was not as clear. Weak Rh\textsubscript{D} (D\textsuperscript{w}) variants bound only 10–15 per cent of the antibody that could com-
bine with other Rh (D) positive erythrocytes, but four examples of Rh (D) positive cells not obviously weak variants (D'), failed to bind a significant amount of tagged antibody. The data reveal considerable evidence of the heterogeneity of Rh antibodies and of the mosaic nature of the Rh (D) antigen.—R. E. R.

**Specific Inhibition of Rh (D) Antibody by Sialic Acids.** M. C. Dodd, N. J. Bigley, and V. B. Geery, Columbus, Ohio. Science 132:1398–1399, 1960.

The agglutination activity of anti-Rh (D) serum with trypsin treated red cells, was inhibited with large concentrations of N-acetyl neuraminic acid (NANA). The effect was noted at pH 3.0–7.2 and was reduced after five weeks refrigerated storage. Beef brain ganglioside containing 17 percent NANA and a *Pseudomonas* polysaccharide were also effective inhibitors, but N-glycoyl-neuraminic acid and D-mannose were weak. All were specific for anti-D and without effect on anti-C, -E, -c and -e, except crude NANA which partially inhibited anti-C and -E. A positive skin test was obtained with bacterial polysaccharide in a rabbit passively immunized 24 hours earlier with anti-Rh (D). Two preparations containing NANA were said to have formed Rh precipitates. Unpublished data were cited concerning inhibition of Rh antibody by human urinary protein after incubation with trypsin or with mumps virus but not after incubation with influenza, NDV or RDE. —R. E. R.


The patient had nine pregnancies, each one of which ended in a miscarriage at the third to seventh month. She belonged to the blood group A, and the Rh (D) factor was positive. Her serum agglutinated all the red cells used in the test (except her own cells) both at 37 C. and at low temperatures. In the presence of complement, hemolysis was observed at 37 C. against random test cells. Further investigations revealed that her red cells were TJ' (−), and the agglutinin in question appeared to be independent of ABO,MNS or Rh system. Therefore, the authors concluded that her serum contained TJ' antibody. The diagnosis was later confirmed by Drs. P. Levine, R. Sanger and J. Dunsford. None of the members of this family, except the patient herself, was found to be TJ' (−).—K. F.


Uncomplicated neonatal hyperbilirubinemia at 61 hours of age, is described in an A₁ child of an A₂ mother, and attributed to A₁ incompatibility. Anti-A₁ in the serum of both mother and child was said to have been lytic in the mother’s serum and accompanied by anti-Le*. Unfortunately, eluates were not prepared, inhibition studies with soluble ABH substances were not done, ABH secretor status was not determined, and minimal hematological evidence for a hemolytic syndrome, such as reticulocytosis, is not mentioned.—R. E. R.


The sites and kinetics of red cell destruction by antibodies were studied in an attempt to delineate the importance of antibody/antigen proportions. Studies in rats involved the use of a heterologous (rabbit) antiserum and in humans involved anti-B isoantibody (passive immunization of an agammaglobulinemic subject with anti-B serum) and passive immunization of several D-negative subjects with various amounts of anti-D serum. The appropriate Cr₁₃₁ labeled “incompatible” erythrocytes were then introduced and the sites and rate of destruction were evaluated. With the incomplete anti-D antibody, moderate amounts caused a splenic pattern of destruction but large amounts caused a moderate hepatic uptake similar to the effects observed with very small amounts of anti-B antibody. With the complete antibodies (human anti-B and rabbit-anti-rat erythrocyte) similar spectra of activity were observed: (1) When small amounts of antibody were employed, slow incomplete sequestration by the spleen occurred. (2) With larger amounts of antibody a rapid, largely hepatic sequestration occurred. (3) With still larger amounts, intravascular lysis was seen, with hemoglobin deposition in liver, spleen, and kidney. The lungs and bone marrow did not participate in the sequestration process at any dose of antibody. The studies show that the differences in in vivo erythrocyte destruction by complete and incomplete antibodies are modified by purely quantitative factors.—H. F.

ABSTRACTS

In vivo experiments in chickens revealed higher uptake of labeled amino acids in Hgb 2 than in Hgb 1, which correlates well with the increasing amounts of the former hemoglobin relative to Hgb 1 occurring during the course of the experiment. In vitro results reveal more uptake in Hgb 1, but the variability of results is probably related to differences in composition or in the state of the internal milieu. Hybridization experiments between Hgb 1 and Hgb 2 support the concept of asymmetrical dissociation. The finding of three varieties of subunits electrophoretically suggested that one of the subunits is common to both hemoglobin types.—A. I. C.

Hemoglobin Peptides Used in Hemoglobin Synthesis. H. Brown and J. Brown. From the University of Wisconsin School of Medicine, Madison, Wis. Metabolism 6:587-593, 1960.

Peptide digests were made of rat and human hemoglobin and the individual peptides qualitatively analyzed for amino acid content by standard techniques. Considerable similarity was noted between the peptide composition of the respective hemoglobins. By using C14 labeled glycine, it was found that radioactivity concentrated preferentially in one peptide. Administration of the labeled peptide to other animals preserved the label in almost one peptide. Administration of the labeled peptide to other animals preserved the label in the same peptide of the recipient's hemoglobin. The use of both countercurrent distribution and separation of the peptides on Sephadex, a dextran polymer which acts as a molecular sieve, permitted quantitation of the amino acid content of at least 13 tryptic peptides. The authors were able to account for almost all the amino acid residues believed to be part of the α-chain. —A. I. C.


Amino acid analyses of the tryptic peptides of the α-chain of human hemoglobin are reported. The use of both countercurrent distribution and separation of the peptides on Sephadex, a dextran polymer which acts as a molecular sieve, permitted quantitation of the amino acid content of at least 13 tryptic peptides. The authors were able to account for almost all the amino acid residues believed to be part of the α-chain. —A. I. C.


The in vivo oxyhemoglobin dissociation curves of 23 anemic patients with Hgb A were determined by measuring oxygen tension, oxygen saturation and pH of the blood while the patients breathed different concentrations of oxygen. A reduction in the affinity of hemoglobin for oxygen was demonstrated in patients with hemoglobin levels below 9 Gm., the displacement becoming more marked as the hemoglobin level decreased below 6.5 Gm. per 100 ml. Correction of the anemia shifted the oxygen dissociation curves back to normal. In patients with the hemoglobinopathies, the abnormality of oxygen affinity seemed to be related to intrinsic defects of the hemoglobin and unrelated to anemia per se. The implication of the reduced affinity of hemoglobin for oxygen in the presence of anemia is discussed and the conclusion reached that the change is a "desirable one because it facilitates the extraction of oxygen from the hemoglobin by the tissues."—A. I. C.


A rapid micro agar gel technic for demonstrating Hgb A2 is described. Additional features of the method permit easy quantitation of Hgb A2, quantitative recovery of the unaltered native protein and the demonstration of several additional unidentified components, particularly in thalassemia. —A. I. C.


The authors report several modifications in the starch-gel technic which improve the accuracy of measuring Hgb A2 levels. These involve a change in the time for boiling the starch (3 minutes) and pouring and cooling of the gel at room temperature. Finally, changes in the photographic recording of results are discussed. Good reproducibility of A2 levels was easily achieved and measurement of the minor component of adult hemoglobin was feasible even in the presence of Hgb S.—A. I. C.

Immunologically Different "Normal" Fetal Hemoglobins. W. F. McCormick and R. H.
Anti-fetal hemoglobin sera were prepared to a single specimen of cord blood containing 91.8 per cent Hgb F as well as to a pooled specimen of several cord bloods. Double diffusion in agar revealed three types of Hgb F on the basis of different patterns of precipitation. The authors suggest that these findings support the concept of heterogeneity of electrophoretically identical fetal hemoglobins.—A. I. C.


The α chains of both Hgb A and Hgb F were found to be very similar on the basis of both “fingerprint” and hybridization experiments.—A. I. C.


Step-wise degradation of the N-terminal portion of the α, β, and γ chains of human hemoglobins A, S and F has been carried out by the Edman technic using phenylthiohydantoin. These studies corroborate the results obtained by Sanger’s DNP method for the first two or three members of the N-terminal sequence and extend the sequence to several additional residues.—A. I. C.


The differential staining of fetal hemoglobin-containing red cells permits recognition of such cells in the presence of cells containing Hgb A or Hgb S. The authors have modified the technic of Kleihauer et al. to improve the differential staining qualities of the Hgb F containing erythrocytes after extracting the adult hemoglobin with acid phosphate buffer. Either an iodination-coupled tetrazonium stain or the ferric ferrocyanide reduction test provides much more striking differences between the pale adult and deeper staining fetal blood cells than was possible with methods previously tested.—A. I. C.


A method is given for staining erythrocytes containing Hgb F in blood smears. In this way it is possible to demonstrate the presence of fetal erythrocytes in the blood of pregnant women as well as the presence of Hgb F-containing RBC in the blood smears of patients with thalassemia or with Hgb S-thalassemia. Blood smears are processed within one hour after preparation and fixed with 80 per cent ethanol for 5 minutes. Hgb A is eluted by a citric acid buffer (pH 3.2) within 5 minutes, and after briefly rinsing with water the slides are stained with hematoxylin (3 minutes) and 0.1 per cent erythrosin (3 minutes). When blood is smeared on a collodion membrane, electron microscopy is possible. In the blood smears of normal adults no RBC containing Hgb F were found. Blood smears from 50 women were investigated during pregnancy and shortly after delivery. In 30 per cent fetal RBC were found during pregnancy and in 70 per cent after delivery. Among 222 blood smears from 118 pregnant women—from the fifth month of pregnancy until shortly after delivery—118 were positive. The number Hgb F-containing erythrocytes was very small; in the most instances there were found only 1–5 per smear (1:5–20 millions). Nevertheless authors were able to demonstrate the accuracy of the blood smear method by mixing normal blood with that known to contain Hgb F and comparing content in the mixtures to chemical methods. In one case, a short time after birth the child suffered from shock and severe anemia. Authors found 90 per cent fetal RBC in the blood smear of the mother. Hgb F erythrocytes were found in patients with congenital spherocytosis and in patients with leukemia. In thalassemia minor there were usually found about 2–5 per cent; in Cooley’s anemia the values were up to 90 per cent or more. It is of interest that in thalassemia minor there were erythrocytes which contained Hgb F and Hgb A.—H. M.


The hereditary persistence of Hgb F in three generations of a Negro family is described. No
hematologic or clinical abnormalities were noted in the three individuals with the isolated “high F” syndrome. Three other members have the combination of Hgb S and F with no demonstrable Hgb A, in spite of proved heterozygosity for Hgb S. Only minor abnormalities from clinical and hematologic standpoints were present in this group. Genetic implications of the high F syndrome are discussed.—A. I. C.


The authors have carried out dissociation and recombination experiments between Hgb A and Hgb S utilizing C14 tagged hemoglobin. Results support the concept that the genetic abnormality in sickle cell anemia manifests itself in an abnormal β chain in Hgb S.—A. I. C.


The authors critically review the literature relative to the renal concentrating defect in sickle cell disease and report a series of experiments in isosthenuric sickle cell subjects observed during their response to water loads, subsequent administration of Pitressin and alterations in filtered solute loads under conditions of maximum hypotension. On the basis of the data reported, the authors suggest that the difficulty in sickle cell disease lies in the ineffective trapping of solute in the medullary circulation of the kidney. They attempt to relate this abnormality to known pathophysiological alterations in the kidney in the sickle cell disorders.—A. I. C.


Studies of the renal concentrating ability in children with sickle cell anemia and its variants are reported. Among the conclusions drawn are that solute free water reabsorption during osmotic diuresis \( (T_\text{m} H_2O) \) was found to be essentially normal in 20 children with sickle cell disease in spite of a definite reduction in maximal urinary osmotic concentration. It is suggested that the “effect of intravascular sickling on renal concentrating power measured by maximal urinary osmotic concentration is mediated through dynamic alterations in intrarenal circulation or by the production of anoxic damage to renal tubular cells.” It seems improbable that an increase in plasma osmolarity plays a role in the vascular occlusive phenomenon in the sickling disorders. The authors discuss possible explanations for the dissociation of the two indices of renal water reabsorption ability.—A. I. C.


A case of homozygous hemoglobin C disease in a 69 year old Anglo-Saxon individual is described. Although the critical genetic data were unavailable in the family study, clinical and hematologic features were consistent with the diagnosis of pure Hgb C disease. Suggestive evidence of a mild hemolytic process was obtained, but no intravascular hemolytic crystals were found.—A. I. C.


Hereditary nigremia, a congenital disease characterized by cyanosis of the mucous membranes and skin, found in Iwate Prefecture, was re-examined from the viewpoint of hemoglobinopathy. Conventional techniques for the detection of abnormal hemoglobin gave normal results. However, with agar-gel electrophoresis and Amberlite IRC 50 chromatography, the hemolysate of nigremia demonstrated a new abnormal hemoglobin. The abnormal hemoglobin was isolated as a dark, greenish brown, thick band in agar-gel electrophoresis and as a similar layer in Amberlite IRC 50 chromatography. Eluate prepared from the abnormal band and subjected to spectroscopy revealed that the abnormal hemoglobin was identical to hemoglobin M. It was tentatively designated as hemoglobin M_1.—K. F.


Hemoglobin H is reported to be a polymer of four β chains on the basis of both "fingerprints" and hybridization experiments.—A. I. C.
HEMOGLOBIN H-THALASSEMIA DISEASE IN A SEPHARIC JEWISH FAMILY FROM TURKEY. Ch. Klibansky, M. Djaldetti, H. Joshua and A. De Vries. From the Hematology Clinic, Department of Medicine D, Rogoff Medical Research Institute and Laboratory of Clinical Pathology, Beilinson Hospital, Petah Tiqwa, Israel. Israel Medical Journal. 19: No. 7-8, 1960.—B. R.


C14 heme and globin were measured in three patients with thalassemia and two normal individuals after the administration of C14 labeled glycine. The results suggest that, in patients with thalassemia, less glycine is utilized for hemoglobin synthesis at a rate slower than normal while fecal stercobilin labeling is extremely rapid. The relative labeling of heme and globin (heme: globin ratio) is significantly lower in thalassemia than in normals. "These results are compatible with the concept that there is a defect in hemoglobin synthesis as well as an abnormality in erythrocyte production in thalassemia major. The enhanced early labeling of stercobilin implies an abnormal degree of premature destruction of red cells or hemoglobin within the bone marrow, or an anabolic pathway for the production of stercobilin. The basic abnormality would seem to be in the erythroblast, a genetic biochemical lesion, it is suggested, not yet identified."—A. I. C.

HEMOSTASIS


Elastase has a blood coagulation-promoting effect. It converts prothrombin to thrombin, but does not act on pure fibrinogen. The effect of elastase on blood coagulation resembles that of trypsin.—S. R. H.


Five cases with neoplastic disease (four cases of prostatic and one of pulmonary neoplasm) were shown to have a cold precipitable plasma protein that migrated electrophoretically like fibrinogen and was clotted by thrombin. The sera had no cold precipitable protein. The cases exhibited hemorrhagic tendencies and perhaps increased tendency to intravascular thrombosis. In vitro tests revealed that other coagulation factors came out with the cryofibrinogen when the latter was separated from plasma in the cold and washed with cold saline. The authors suggest the theory of Henstell and Kligerman, as an explanation for the appearance of both intravascular clotting and hemorrhagic diathesis. The complexing of coagulation factors with the cryofibrinogen might favor local thrombosis when the cryofibrinogen is precipitated locally. If the precipitates were large in amount and filtered from the circulation in the capillary bed it might result in hypocoagulability and hemorrhage.—R. G.


In rabbits an antihistaminic drug, Pernovin, in daily doses of 4 mg./Kg., body weight improves blood coagulation disorders following total body irradiation (1700 r). It inhibits the appearance of labile fibrinogen. Author assumes that this result proves the primary role of histamine liberation in the development of postirradiation defect of coagulation.—S. R. H.


A case of severe factor X deficiency is reported. The patient was a 27 year old female with markedly prolonged prothrombin and stypven times, prolonged clotting time and poor thromboplastin generation. During pregnancy there was a marked reduction in the prothrombin time and the thromboplastin screening test and partial thromboplastin test became normal. The improvement began within one to three weeks following conception and continued for at least one week after delivery. Bleeding tendencies stopped during pregnancy and, based on history of her previous pregnancies, improvement persisted during the period of lactation.—R. G.

ABSTRACTS

Of 81 patients with proved acute myocardial infarction, 41 were treated with heparin and 40 received no anticoagulant therapy. Heparin was administered intravenously every six hours through an indwelling venous catheter. Heparin dosage varied between 50 and 150 mg, the dosage being adjusted to obtain clotting times of two to three times the control value 3½ hours after the preceding dose of heparin. There was no evidence from this study that heparin was beneficial in reducing the incidence of any complications during convalescence from acute myocardial infarction. Definite thromboembolic episodes occurred with similar frequency in both groups. The total number of mural thrombi, arterial emboli, thrombophlebitis in lower extremities, pulmonary emboli and recurrent myocardial infarction was 10 in the heparinized and 12 in the control patients.—R. G.


A survey of the results of various forms of therapy in 744 cases of venous thromboembolism is reported. According to these findings, heparin is the anticoagulant drug of choice and the best results are achieved when it is administered by the intermittent intravenous method. The authors recommend a dosage schedule of 75–100 mg every 6 or 8 hours and recommend that the clotting time should be doubled 2 hours after a dose of heparin. They found thatbishydroxycoumarin was much less effective from the standpoint of prevention of pulmonary embolization, prevention of recurrence of the thrombotic process or speed of subsidence of the inflammatory process. Intravenous heparin afforded the best protection against pulmonary embolization and recurrence of thrombophlebitis when compared to subcutaneous or intramuscular heparin, coumarin therapy, or heparin plus coumarin therapy. The authors feel that pulmonary embolization is an indication for venous interruption which must be followed by heparin to prevent thrombosis and embolism from arising proximal to the ligation. The mortality of pulmonary embolism in the series was 26.5 per cent. The mortality for cases in which there were sufficient warning signs of pulmonary emboli for therapy to be instituted was 40 per cent.—R. G.


A patient is described who had been satisfactorily controlled on long term anticoagulant therapy (bishydroxycoumarin) for over two years, and who, without evident cause, suddenly presented signs of an acute surgical abdomen. At operation, massive hemoperitoneum was found and the cause was identified as bleeding from a ruptured corpus luteum.—R. G.


Severe dermatitis involving the skin and mucous membranes followed the use of sodium warfarin for treatment of cerebral thrombosis. The lesions disappeared with steroid therapy but recurred when warfarin was again tried some 10 weeks later. Lesions again cleared when medication was withdrawn. Warfarin applied to the skin caused erythema. Of interest is the fact that the patient developed no skin lesions or other allergic manifestation when bishydroxycoumarin was used either before or after the use of warfarin.—R. G.


A case of acute leukemia with a lethal hemorrhagic syndrome due to fibrinolysis and thrombocytopenia is presented. The importance of the fibrinolytic mechanism in the production of severe bleeding during acute leukemia is emphasized. The interesting relation between fibrinolysis and the peroxidase positive promyelocytes of those cases is discussed.—B. R.


Ultraviolet irradiation of mice is followed by marked thrombocytosis for 14 to 16 days, with a peak on the sixth day. In parabiotic mice thrombocytosis also develops in the animals which were protected from irradiation. Serum taken 24 hours after irradiation induces thrombocytosis when given to normal rats. Serum taken at the peak of thrombocytosis is less effective. The active serum is sensitive to heat and trypsin. The thrombopoietic serum factor is possibly of protein character.—S. R. H.

Rabbits given intravenous injections of 15–20 ml. of 2 per cent India Ink solutions (400 to 500 mg. of ink) exhibited an abrupt drop in platelet count levels from normal levels of 138,000 to 750,000 per mm² to 1,000 to 12,000 per mm². Thereafter the count gradually returned to normal levels within two to four hours. By means of transfusions of in vivo 3H² labeled platelets into recipient animals it was possible to show that the disappearance of platelets from the circulating blood after administration of India Ink was not due to destruction. The recovery of the platelet count was not accomplished by the release of a new generation of platelets but by the release of the sequestered platelets, since there was no fall in the specific activity of the platelets. Observations on blood smears made some hours after India Ink showed groups of agglutinated platelets covered with India Ink particles, and the histologic preparations of lymph node, liver, bone marrow, and spleen showed agglutinated India Ink particles with huge numbers of platelets. When prelabeled blood of donor animals which had received injections of India Ink was transfused into recipient animals 17–20 hours after administration of ink, only a small proportion of the radioactive platelets could be recovered from the recipient animal. This suggested that the transfused platelets had been damaged by the ink and had been removed and destroyed by the recipient animal.

—R. G.


Platelet counts (Brecher-Cronkite method) in venous blood and in capillary blood from deep cuts of the skin were identical. Platelet counts in blood from a standardized, superficial cut on the forearm were significantly lower than those of venous blood. The difference is believed to be an in vivo measure of platelet adhesiveness. The lower platelet count in blood from superficial cuts could not be attributed to dilution with tissue fluid because the hematocrit was almost the same as in venous blood, nor was it due to coagulation, as it could be demonstrated in heparinized patients also. In 50 normal individuals the number of adhesive platelets (difference between platelet count in venous and capillary blood from superficial cut) was 96,000 ± 24,000 per cu. mm. or 37 ± 7.5 per cent of the total count. The corresponding figure in a patient with thrombasthenia was 3,000 or about 3 per cent of the total platelet count.—S. A. K.


Platelets were counted automatically in an electronic cell counter ("Celloscope"). One milliliter of venous blood is drawn into a syringe with 1 ml. of 3.8 per cent sodium citrate. The contents of the syringe are immediately mixed with 18 ml. of a glass-filtered diluting fluid (930 ml. of 0.9 per cent NaCl, 50 ml. of M/15 phosphate buffer at pH 7.55, 10 ml. of 1 per cent EDTA solution, and 10 ml. of a 35 per cent formaldehyde solution, the pH of which has been adjusted to about 8.0). The mixture is left for sedimentation. After a few hours 0.05 ml. of the supernate is removed from about 1 cm. below the surface in the axis of the tube, and diluted with 20 ml. of the diluting fluid. A sample of this is now counted twice. The first count is done with the instrument set at high sensitivity, i.e. including small particles in the count, to register platelets as well as red cells. The second count is done at a setting which counts the red cells but disregards the platelets. The difference between the two counts is the platelet count. The coefficient of variation (10 counts of one sample) at a mean platelet count of 335,600 was 4 per cent. At 26,100 platelets it was 12 per cent. In general, the coefficient of variation in hemocytometer counts was about twice that of the automatic counts. A crucial point in this method is the sedimentation rate of the platelets. Under the experimental conditions, no significant sedimentation of platelets occurred during the first six hours.—S. A. K.

Miscellaneous


Using specific antisera against purified proteins in immuno-electrophoresis and in Ouchterlony double diffusion methods, the author has convincingly demonstrated the presence in normal urine of proteins immunologically identical with the plasma proteins pre-albumin, albumin, ceruloplasmin, transferrin and gamma globulin.—H. F.

Isolated Cells: Normal and Tumor. I. Preparation and Protein Synthesis of Thymocytes

ABSTRACTS
ABSTRACTS


Isolated mammalian cells are more satisfactory for many metabolic studies than are tissues or tissue slices, particularly for problems involving cell permeability and intracellular metabolic processes. The authors describe a method whereby individual cells may be separated from the thymus tissue of rats and from rat ascites tumors. Cells isolated by this technic continue to synthesize protein in vitro for at least four hours, during which time viability is retained, as indicated by the lack of permeability to eosin Y. Incorporation of C14-labeled leucine, valine, glycine, and alanine proceeded at a rate equal to that obtained in tissue slices. The second paper applies this method to studies of the effects of non-ionic detergents on normal and neoplastic cells; these depressed both protein synthesis and the intracellular concentration of amino acids, presumably due to effects on the cell surface. The methods outlined in these two papers should find ready application in the study of leukemic cells.—H. F.


A macroglobulin $S_{20,7}^{w}$ 17 with a molecular weight of 1,200,000, and a relative concentration of 13 per cent of total proteins was found in a patient with plasmocytoma. Electrophoretic analysis revealed that the macroglobulin migrated very near to the gamma-globulins. An abnormal fraction was obtained from an extract of lymph node and presented an electrophoretic mobility which was similar to that of the abnormal serum fraction. Immunological investigations by means of agar-gel diffusion confirmed the similarity of the two fractions. A lymphoglandular origin of these macroglobulin was postulated on the basis of the results obtained.—P. D. N.


This is a study of 18 patients with macroglobinemia—defined here as a condition in which more than 10 per cent of the serum proteins have a sedimentation value greater than $S_{20,7}^{w}$ 16. Symptoms and signs in these patients were varied and, apart from striking fundal changes, non-specific; limited observations on biopsy and necropsy material showed normal appearances or diverse pathological changes from infiltration with plasma cells or lymphocytes to the features of a reticulosis, and the author discusses the relation of the condition to myelomatosis and to the reticuloses. Immunological studies on purified macroglobulin from patients did not show any difference between this and macroglobulin from normal serum; further, the amino acid composition was not strikingly different from that of normal $\gamma$-globulin. In view of these findings, the diverse clinical and histopathological features, and the demonstration of macroglobulins in small quantities in normal subjects—and in excess in patients with liver disease, syphilis, and chronic infections—the author does not think that there is any justification for regarding macroglobulinemia as a nosological entity.—R. M. H.


This paper gives the case report of an elderly man with macroglobulinaemia and hemolytic anaemia. Splenectomy resulted in partial remission of his anaemia. Histological examination of the spleen and of lymph nodes showed evidence of marked erythrophagocytosis.—C. C. DE G.


The presence of multiple separable serum factors reactive with various components of cell nuclei has previously been demonstrated in sera of patients with lupus erythematosus and other diseases. The present paper deals with characterization of these factors as 7S or 19S gamma globulins. The serum factor responsible for LE cell formation and the factor active with calf thymus nucleoprotein were both found only among the 7S gamma globulins, whereas the factor with serologic activity (tanned cell hemagglutination method) directed against human liver nucleoprotein was found mainly, although not exclusively, in the 19S gamma globulins. The activity
against cell nuclei detected by the fluorescent antoglobulin technic was found with the 7S gamma globulin in those LE sera reactive with calf thymus nucleoprotein, but was found with both the 7S and 19S gamma globulins in LE sera reacting primarily with human liver nucleoprotein.—H. H. F.


Complications after long-term corticosteroid therapy in 99 patients out of 262 suffering from different blood diseases are described. 1. The most frequent were purulent and infectious complications (20 per cent), gastric and intestinal disturbances (10 per cent) circulatory (6 per cent) hormonal (5 per cent) and neurologic (2 per cent). 2. Complications were encountered more often in patients suffering from leukemia or pancytopenia. 3. There was a relationship between dose and incidence of complications: 56 per cent in patients who had received large doses of the drug.—E. K.

**Erratum**

The Editorial Board page in the February, 1961 issue was inadvertently printed from 1960 plates and hence was incorrect, for which apology is made. The correct listings are printed in this issue.
ABSTRACTS