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ERYTHROCYTES


Less time consuming in vitro methods of erythropoietin assay based on the incorporation of 14C-glucosamine and 59Fe into marrow cells have been compared to the exhypoxic polycythemiac mouse assay. Erythropoietins from various sources, including sheep and human plasma as well as human urine, were compared after proper dilution to eliminate the effect of inhibitors present in the different preparations. Their specific activities varied widely depending on the assay system employed. To account for these differences the authors favor the hypothesis that in erythropoietin preparations several different biologically active entities may coexist. It is suggested that erythropoietin action results from the sum total of the processes regulated by a number of factors. Thus the activity of an erythropoietin preparation measured in vivo reflects its content of the factor present in rate-limiting amounts required for the complete development of red cells. On the other hand the in vitro tests reflect the content of factors stimulating specific processes such as heme synthesis or glucosamine incorporation into macromolecules.—M.S.

ERYTHROPOIETIN INHIBITOR IN PLASMA FROM PATIENTS WITH CHRONIC RENAL FAILURE. Y. Moriyama, H. Saiito, and Y. Kinoshita. Second Department of Medicine, Niigata University School of
DUODENAL FERRITIN SYNTHESIS IN IRON-REPLET AND IRON-DEFICIENT RATS: RESPONSE TO SMALL DOSES OF IRON. G. M. Brittin and D. Bacal. The Department of Clinical Pathology, The University of Texas at Houston, M. D. Anderson Hospital and Tumor Institute, and the Department of Clinical Pathology and Laboratory Medicine, University of California Medical Center. J. Lab. Clin. Med. 77:54-58, 1971.

The authors previously demonstrated that administration of pharmacologic doses of iron stimulated ferritin synthesis in the duodenum of iron-deficient rats and that this newly synthesized ferritin did not prevent increased iron absorption from the intestine. In the present study it could be shown that both iron-replete and iron-deficient animals incorporated 14C-DL-leucine into duodenal ferritin in response to the intraduodenal instillation of small doses of iron ranging from 10-500 µg. The incorporation of leucine into the immunologically isolated ferritin was linear for iron-deficient rats over the entire dose range studied, but this was not the case for the iron-replete animals. The finding that small doses of iron, which are known to be rapidly absorbed, induce duodenal ferritin synthesis in iron deficiency provides evidence against the hypothesis that it is the role of intestinal ferritin to prevent the absorption of iron by binding it in the mucosa. The authors propose that the observed intestinal ferritin synthesis in iron-deficient rats may represent a cellular defense mechanism against the well-recognized toxic effects of ionic iron.—M.S.


Among 19 U.S. Marines with malaria in Vietnam, all of whom were receiving treatment with pyrimethamine, 18 had minimal to moderate megaloblastic changes of the bone marrow, 13 had leukopenia and 13 had a hematocrit value of < 35%. Megaloblastic changes were also present in four of ten subjects with other disorders and none of them were anemic. Serum vitamin B12 concentrations were consistently normal, but serum folate concentrations were subnormal in 17 patients with malaria and in seven of the other ten subjects (in
three of whom values were 0). *Abstractor's comment:* Whereas the reduced folate concentrations and hematologic changes are probably attributable to pyrimethamine in the patients with malaria, the high prevalence of low folate values in the other subjects is surprising and unexplained.—F.A.K.

**ADRENOCORTICAL FUNCTION AND THE EFFECT OF CORTICOSTEROID TREATMENT IN ADDISON-BIERMER DISEASE.** W. Hartwig, S. Paucelsk, B. Migdalaka, R. Rechovies, and A. Rociszewska. First Department of Internal Medicine, Postgraduate School of Medicine, and the Department of Internal Medicine, Institute of Hematology, Warsaw, Poland. Acta Haemat. Pol. 1:61–68, 1970.

Numerous tests of adrenocortical function were performed in ten cases of pernicious anemia showing no essential abnormalities. However, treatment with prednisone during 2–3 mo resulted in nine cases in a 3-10-fold increase in absorption of vitamin B₁₂ independently of the presence or absence of antibodies against intrinsic factor. This improvement lasted 2–3 mo in all cases but in one case increased absorption of B₁₂ persisted for over 6 mo after prednisone withdrawal.—M.K.


Red blood cell (RBC) survival was studied in ten patients with aplastic anemia due to different causes by using ⁵¹Cr-labeled autologous RBC. The following observations were made: In six patients with idiopathic aplastic anemia in which the RBC half-life ranged from 6 to 23 days there was correlation between the degree of shortening in RBC survival and the time of patient survival. Such correlation was not found in other four cases, which included drug-induced aplastic anemia and osteomyelofibrosis.—Z.R.

**LEUKOCYTES**


The present study concerns the relative frequency of reticulum cell sarcoma, lymphosarcoma and Hodgkin’s disease in Japan, the United States, and Great Britain. Application of Western diagnostic techniques to Japanese case material suggests that a portion of the apparent differences between East and West in this respect may be due to discrepancies in classification and/or histologic interpretation but that not all such differences can be explained on this basis. Reticulum cell sarcoma is the most prevalent form of malignant lymphoma in Japan with a relative frequency (42%) that approaches Hodgkin’s disease in Western series (49%). Conversely, Hodgkin’s disease is the least frequently encountered form of lymphoma in Japan, with a relative frequency (20%) not far removed from reticulum cell sarcoma (18%) in the West. There is little difference in relative prevalence of lymphosarcoma in Japanese and Western experience. Finally, each type of lymphoma in Japan appears to be associated with a shorter estimated clinical course than the comparable disease in the United States. It is suggested that these geographic differences may be related to the close interrelationships of the lymphomas and genetically governed variations in host reactivity.—J.E.U.


Since deoxyribonucleotides exist in very low concentrations in cells, ribonucleotide reductase may function as a rate-limiting enzyme for the formation of DNA. Of the two reductases which have been purified from bacteria, *E. coli* and *L. leichmannii*, the enzyme isolated from human bone marrow resembles that obtained from *E. coli* in respect to its substrate specificity (that is, ribonucleoside diphosphates) and its independence from B₁₂ coenzyme. This enzyme was found present in leukocytes from...
blood of patients with acute or chronic myelocytic leukemia. The specific activities were similar to those found in normal bone marrow. Lymphocytes from patients with chronic lymphocytic leukemia and normal peripheral leukocytes had much lower specific activities. There was a 20-fold increase in ribonucleotide reductase activity in megakaryoblastic bone marrow from patients with B12 deficiency as compared with normal bone marrow. The mechanism underlying this striking elevation of the enzyme activity is still uncertain.—M.S.


The frequency of nine HLA leukocyte antigens in two prospective series of patients with Hodgkin’s disease (35 and 75 patients) has been determined. An antigen previously known as 4c was found in 56% and 45%, respectively, of the patients in the two series compared with 25% in the normal Australian population. This difference appears to be due to an increased frequency of an included antigen, W5. Family studies have shown a normal segregation of antigen W5, but have not as yet revealed any abnormal frequency of haplotypes. It is suggested that genetic or viral factors may be significant in the pathogenesis of this disease.—J.E.U.

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The uptake of 14C-cortisol by leukemic cells was measured in vitro using cells from patients with chronic lymphatic leukemia, chronic granulocytic leukemia, and acute myelogenous leukemia, as well as cells of L1210 mouse leukemia. Leukemic cells were suspended in Hanks’ solution and patient’s serum, and were incubated with 14C-cortisol at 37°C or 0°C. The uptake of cortisol by leukemic cells was rapid during the initial 15 min and became gradually slower, reaching a constant value after 60 min of incubation. The uptake was higher at 37°C than at 0°C. An uptake proportional to the concentration of cortisol was observed with either incubation temperature. No difference in uptake due to temperature differences was noticed at pH 6.5 of the medium. The highest uptake was seen at pH 6.5–8.0. The distribution of cortisol in subcellular fractions of leukemic cells after incubation of 30 min was 14.5% in the nucleic acid, 9.3% in protein, and 76.2% in the acid-soluble fraction using Schneider’s preparation. The amount of cortisol in the nucleic acid fraction was constant during more than 30 min incubation. Serum in the incubation medium exerted an inhibitory effect on the uptake of cortisol. The uptake of cortisol by various leukemic cells was different in the different types of leukemia.—K.F.


The work was aimed at the isolation of the whole histone protein and of the four main histone fractions from human normal and leukemic leukocytes from peripheral blood, and at the measurement of their 14C lysine incorporation rate. Isolated proteins were identified by starch and acrylamide gel electrophoresis and by amino acid analysis. The obtained results seem to suggest that although the relative rate of synthesis of the whole histone protein is different in various types of leukocytes, the general pattern of histone fractions synthesis is similar in all types of leukocytes at least as far as fractions F1, F2a and F2b are concerned.—M.K.

Seventy-two patients with Hodgkin's disease, reticulum cell sarcoma, lymphosarcoma, and various other malignant neoplasms were treated with the methylhydrazine derivative procarbazine hydrochloride (Matulane, NSC-77213). Response to therapy could be evaluated in 50 patients. Favorable responses occurred in 22 of 33 evaluable patients with Hodgkin's disease, two of five patients with reticulum cell sarcoma, and two of five patients with lymphosarcoma. All of the responders had previously shown resistance to at least two other antitumor agents. Toxic effects of therapy included leukopenia, thrombocytopenia, nausea, vomiting, stomatitis, lethargy, ataxia, and alopecia. Median time to start of response was 21 days; median cumulative dose to start of response was 51.3 mg/kg. Responses lasted a median of 98 days (range: 34–322 days). Procarbazine is a useful agent in advanced lymphomas.—J.E.U.


Forty patients who had acute leukemia received therapy with cytosine arabinoside and cyclophosphamide. The response rate was 53%. Sixteen of the 34 patients (47%) who received an adequate trial of therapy achieved a complete remission. The response rate was 73% for patients less than 35 yr old, 36% for patients 36–65 yr old, and 67% for patients over 65 yr old. The response rate was 41% for the 29 patients who had acute myelogenous leukemia and 82% for the 11 patients who had acute lymphocytic leukemia. The median survival time for responders was 7 mo. The most common toxic effect was myelosuppression, although nausea, vomiting, alopecia, and hemorrhagic cystitis were noted.—J.E.U.

TREATMENT RESULTS WITH VINBLASTINE IN MALIGNANT LYMPHOPROLIFERATION. P. Klener, L. Donner, and R. Neuwirtová. The Department of Medicine, University Charles, Prague, Czechoslovakia. Vnitřní Lek. 16:638–643, 1970.

A group of 19 patients with the generalized form of Hodgkin's disease were treated with vinblastine doses of 0.1–0.3 mg/kg/wk intravenously. Eighteen patients had been treated with various cytotoxic drugs or with radiation therapy before vinblastine was started. The medication was given for a mean period of 42 mo. The treatment failed in three patients who were in the terminal phase of the disease. Remission was obtained in ten patients (52.6%) who received short-term therapy and lasted for an average of 10.2 wk. In six patients (31.5%) complete remissions were achieved which are still continuing after more than 3 mo. All patients tolerated the administration of vinblastine very well. Leukopenia was recorded in five patients; in three of them it was only temporary.—L.D.


The authors compared the life-span of 137 adult patients with acute leukemia who were hospitalized at the Second Medical Department in 1934–1948 (without specific treatment) with that of 81 patients hospitalized in 1951–1967 (treated with corticoids and cytostatic preparations). The period of survival in the latter group was significantly higher and was on the average 5% mo as compared with 1 mo in the group without specific therapy.—L.D.


A 53-yr-old man with untreated acute myelomonocytic leukemia was awakened from sleep in his hospital bed by severe epigastric pain, which led to laparotomy. A grossly disrupted spleen was removed; section showed leukemic infiltration of the pulp and capsule, with many hemorrhages. This is only the ninth case of genuinely spontaneous rupture of the spleen recorded...
in acute leukemia: all others had been subjected to varying degrees of trauma. Any patient with leukemia and otherwise unexplained abdominal pains should be suspected of having a ruptured spleen.—F.W.G.

STEM CELL LEUKAEMIA IN MYELOMATOSIS.


Four patients with myelomatosis of long duration developed, as a late phenomenon, acute leukemia, with typical myeloblasts in bone marrow and peripheral blood. They had been treated with nitrogen mustard derivatives: two with melphalan, one with cyclophosphamide, and one with both drugs. No other common factors with a hypothetical leukemogenic effect could be demonstrated. It could be that termination of myelomatosis in acute leukemia is a not uncommon, but till now overlooked, phenomenon.—J.E.U.

THE HYPEREOSINOPHILIC SYNDROME.


Symptoms and signs of this disease are described in two women. The common sign of their illness was an increase in the eosinophilic leukocyte count. In the first patient there was hepatomegaly and splenomegaly, together with enlargement of lymph nodes. In the plasma electrophoresis serum beta and gamma globulin were increased. In the second patient there were transitional pulmonary infiltrations and the gamma globulin was only slightly increased. In both cases the illness was of long duration and ended with remission.—L.D.

SUBACUTE MYELOFIBROSIS.

D. Mrkos. The First Medical Department, Medical School of the University of Brno, Czechoslovakia. Vnitrní Lek. 16(10):1066–1073, 1970.

The clinical and pathologic picture of myelofibrosis is analyzed. Only in two out of 10 cases was the correct diagnosis of subacute myelofibrosis made during life, while in the rest the diagnosis was made at autopsy. The clinical picture was characterized by a malignant course and the blood examination suggested atypical blastic leukemia. The decisive means for the correct diagnosis of subacute myelofibrosis is the bone marrow biopsy. The most important difference between the chronic and the subacute form of myelofibrosis is in the different type of extramedullary hematopoeisis, which in the subacute form has signs of dedifferentiation or of maturation inhibition.—L.D.

HEMOSTASIS

VASCULAR LESIONS: POSSIBLE PATHOGENETIC BASIS OF THE GENERALIZED SCHWARTZMANN REACTION.


Evidence is presented for the hypothesis that the generalized Shwartzman reaction is primarily the result of a vascular reaction with in situ formation of thrombi at areas of endothelial cell detachment and exposed basement membrane. After a single small dose of E. coli endotoxin, 80% of the treated rabbits developed circulating endothelial cells (identified by light and electron microscopy), leukopenia, and thrombocytopenia. Fifty per cent of the endotoxin-injected animals presented evidence of endothelial lesions in three randomly selected areas of their aortas. The lesions which were considered positive consisted of separation of endothelial cells from their basement membranes, lytic areas in their cytoplasm, or the presence of large vesicles filled with amorphous material. Prior anticoagulation with heparin did not prevent the endotoxin-induced changes. None of the control animals showed these abnormalities. Based on these results the authors advance the hypothesis that endotoxin is directly toxic to vascular endothelium or exerts its effect through leukocytes. The consequent damage to the endothelial cells leads to their detachment, to exposure of basement membrane, and to platelet accretions at such thrombogenic sites. In the otherwise untreated animals there is additional initiation of clotting and deposition of fibrin.—M.S.

For preparation of fibrinogen from plasma, the Ware, Guest and Seegers freezing–thawing method is considered to be theoretically excellent. However, the method has rarely been adopted because of its very low reproducibility. The authors first established an improved freezing–thawing method with high degree of reproducibility. The product can be stored in plasma, the first established an improved freezing of its very low reproducibility. The authors lowers: (1) Whereas prothrombin was con-plasma by this new method, which can be 94–fled fibrinogen with a clottability of without any change in activity. The pun-in the frozen state for more than 1 yr was done gradually at room temperature. It also eliminated by adapting freeze-thawing again to the final fibrinogen solu-

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respiration by 50% (Crabtree effect) and stimulated glycolysis by 253%. Glycogenolysis was greatly inhibited in this condition. It was also found that under aerobic conditions respiration accounted for 53% of the total ATP production when glucose was present in incubation media and for almost 90% without glucose. It was concluded that respiration represents a very important source of energy in rat blood platelets.—Z.R.


Rats were exposed to a 35–37°C temperature for 48 hr, for 96 hr, or for several months, and the changes in blood coagulation and fibrinolysis were determined. Only in rats adapted to the high temperature of 35–37°C for several months were changes in hemostasis noted. Prothrombin time was significantly prolonged, factors II and V decreased, and the prothrombin lag time shortened. Thrombin time, factor VII, factor X, and TGT did not change. It was suggested that these findings might reflect enhancement of the continuous physiological process of intravascular coagulation.—Z.R.


In this study the contribution of respiration and glycolysis in total energy production of rat platelets was determined in vitro. The rate of respiration was measured by oxygen consumption and the rate of glycolysis by glucose consumption, lactate production (Maitra, P. K., and Estabrook, R. W.: Anal. Biochem. 7:472, 1964) and glycogen depletion (Pfeiderer, G.: Methods of Enzymatic Analysis. New York, Academic, 1965). It was found that presence of glucose in the incubation media inhibited respiration by 50% (Crabtree effect) and stimulated glycolysis by 253%. Glycogenolysis was greatly inhibited in this condition. It was also found that under aerobic conditions respiration accounted for 53% of the total ATP production when glucose was present in incubation media and for almost 90% without glucose. It was concluded that respiration represents a very important source of energy in rat blood platelets.—Z.R.


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months. The inhibitor was not species-specific. With the aid of chromatography on Sephadex G-200 it was isolated in the fraction containing mostly IgG globulins.—L.D.

THE INFLUENCE OF STREPTOKINASE UPON SOME FUNCTIONS OF BLOOD PLATELETS.


The in vitro influence of streptokinase upon some functions of the blood platelets was studied. The concentrations used were 100–2000 units/ml plasma. The streptokinase decreased retention and spreading of the platelets on glass beads and glass slides. It was shown that it could accelerate platelet aggregation, but to a significantly lesser degree than with adenosine diphosphate. Platelet aggregation could not be prevented by substances known to inhibit ADP-induced platelet aggregation. Streptokinase caused a delay of the viscous metamorphosis and of the release of platelet factor 3, whereas the antiheparin platelet factor 4 remained uninfluenced.—L.D.

A CASE REPORT OF INBORN FIBRIN-STABILIZING FACTOR (FACTOR XIII) DEFICIENCY.


This is a case report of a 30-yr-old male with factor XIII deficiency. Extensive hemorrhages in soft tissues, muscles, and joints were the main findings. Diagnosis was established on the basis of the urea solubility test and confirmed by the cross-matching test with plasma from other previously detected cases of factor XIII deficiency in Basel and Belgrade. Parents or other relatives were not available for examination. Abstractor’s comment: This is the second case so far reported in Yugoslavia. The patient recently died with signs of intracranial bleeding, which seems to be a frequent cause of death in this disease.—Z.R.

THE EFFECTS OF CERTAIN SYMPATHOMIMETICS AND SYMPATHOLYTICS ON THE INCIDENCE OF EXPERIMENTAL VENOUS THROMBOSIS.


Thrombosis of the jugular vein was induced in rats by a damage to the vascular wall induced by formaldehyde. In 46% of normal animals, thrombosis developed within 24 hours. Single i.p. injections of adrenaline (0.1 mg/kg body wt) or noradrenaline (0.2 mg/kg body wt or 0.02 mg/kg body wt), given 30 min before the application of formaldehyde, increased the incidence of experimentally produced thrombosis. After the injection of adrenaline, thrombosis developed in 73% and 86.6% and after noradrenaline in 80.6% and 73.3% of rats respectively. Propranolol administered 30 min in advance reduced the incidence of thrombosis following the application of adrenaline to 53.3%; phentolamine reduced it to 60%. The recalification, cephalin, and prothrombin time values were not changed after adrenaline injection. The number of blood platelets and the fibrinogen level were rising. These changes were manifest in animals both with or without thromboses.—L.D.

CHANGES IN BLOOD COAGULATION IN STENOTIC HEART DISEASE.


A series of T.E.G. tests with blood from individual heart chambers in stenotic patients was studied. Blood was obtained by heart catheterization. The results disclosed that the transition of blood through the stenotic orifice markedly increased its readiness for clotting and this was observed also in cases which were on heparin therapy. The increase of the clotting tendency seemed to be in direct relationship with the degree of the stenosis.—L.D.

BLOOD COAGULATION IN PATIENTS WITH RENAL INSUFFICIENCY TREATED BY PERITONEAL DIALYSIS.

Complex tests of blood coagulation were performed in 21 patients with renal insufficiency and repeated in ten of them during treatment with peritoneal dialysis. The most frequent pathologic finding was a hypercoagulative state with an unusual form of T.E.G., hyperfibrinogenemia, and a disturbance in the consumption of prothrombin. In contrast to the result of the consumption test were the normal values of blood platelets. Clinically and biochemically, effective treatment by hemodialysis had no influence on blood coagulation changes.—L.D.

**IMMUNOHEMATOLOGY**


Suspensions of PHA-stimulated human blood lymphocytes were irradiated with X-ray doses ranging from 500 to 5000 R, and after three days of incubation the following parameters were estimated: (1) percentage of blast transformation; (2) diameter of cell nuclei; and (3) incorporation of thymidine-$^{14}$C by the cultured lymphocytes. It was found that the percentage of blast transformation in irradiated cultures was similar to that of non-irradiated controls and was not dependent on the dose of radiation. The diameter of cell nuclei in irradiated cultures was greater when compared to the one in non-irradiated PHA-stimulated controls. With the increase of the X-ray dose the number of cells with expanded nuclei also increased. Although in all irradiated cultures the incorporation of the DNA-precursor into the cells was reduced, this reduction was found to be dose-dependent. At the maximum dose of irradiation (5000 R), 20% of the cells still preserved the ability to incorporate the DNA precursor. From these data a protective effect of PHA on lymphocytes in culture was postulated. It is also suggested that initial metabolic changes in PHA-stimulated lymphocytes could play an important role in the protection of these cells from irradiation.—Z.R.


L-asparaginase inhibited blastic transformation as well as thymidine incorporation in cultures of human peripheral lymphocytes stimulated by phytohemagglutinin. The magnitude of the inhibition was similar for both phenomena. Small numbers of lymphocytes underwent blastic transformation despite the presence of L-asparaginase in the medium. The number of such asparaginase-resistant cells appeared higher in the cultures stimulated by PHA than in control cultures. Some preliminary calculations indicated that the two groups may differ also in their capacity to incorporate thymidine. It thus seems that the population of peripheral lymphocytes derived from adult healthy individuals exhibits an intrinsic heterogeneity of response to the immunosuppressive action of L-asparaginase.—M.K.

**Antibodies to Intrinsic Factor in Serum and Gastric Juice of Patients with Pernicious Anemia. Blocking and Binding Antibodies.** K. Wysocki, W. Fenrych, B. Wierusz, and J. Hansz. Third Department of Medicine, Medical School, Poznań, Poland. Acta Immun. 1:63-68, 1970.

Sera and gastric juice from 20 patients with pernicious anemia were studied to establish the frequency of occurrence of intrinsic factor antibodies (I.F.A.). Blocking and binding antibodies were investigated. In part of the cases the antibodies were present both in the serum and in the gastric juice. In some cases they were present only in the serum or in the gastric juice. Binding antibodies were observed only together with blocking antibodies. The frequency of occurrence of circulating I.F.A. did not correlate with the clinical or hematological features. Some correlation
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between clinical features and presence of antibodies in the gastric juice may be postulated in cases in which I.F.A. was more frequently found during relapse than during remission.—M.K.


In a patient with acute myeloblastic leukemia three populations of erythrocytes were isolated. One contained the substance A₁; the second contained substance A; and the third was devoid of detectable substance A. In the A₁ population, normal content of I-H was found while the third population revealed a significant decrease of substance H, which was a rather unexpected phenomenon. In the first (A₁) and third populations no aberrations as far as the antigen I is concerned, were found, while a significant increase of compound i was observed. The described anomalies are examples of complicated phenotypic modifications within the three group systems, hitherto reported only in hematological diseases.—M.K.


Fifty-two consecutive patients with monoclonal gammapathy (MG) included 31 with myelomatosis, three with Waldenström's macroglobulinemia, and four with other malignancies. Thus, in 73% of cases the abnormal immunoglobulin peak signified a malignancy. Of myeloma patients, 26% had second malignancies; 23% of all patients with MG had no apparent malignancies. In seven cases MG was probably present several years before a malignancy was discovered, and hence the finding of the abnormality must always be taken seriously. IgG levels above 2 g/100 ml were strongly correlated with the presence of myelomatosis, those below 2 g/100 ml with the apparent absence of malignancy. The correlation between high monoclonal IgG and myelomatosis was even stronger when there was concurrent suppression of IgA and IgM.—F.W.G.

Miscellaneous


Clinical and laboratory analyses of 49 patients with detectable Australia (Au) antigen (39 cases) or antibodies (10 cases) were performed. In 39 patients, the signs of liver damage were demonstrated. In eight of them acute infectious hepatitis was diagnosed. Ten patients without symptoms or signs of liver damage in spite of detectable Au antigen had immunoproliferative diseases.—M.K.
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