Technical Papers

The Influence of ACTH and Cortisone on Certain Factors of Blood Coagulation

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In both experimental animals and in man a variety of factors has been observed to induce changes in the coagulability of blood. Cannon (*2*) demonstrated that epinephrine, when injected into the experimental animal in small amounts, accelerates clotting, whereas in a large amount it materially delays coagulation. Moon (*9*), Eagle *et al.* (*4*), Howell (*6*), and Jacques and Waters (*7*) have ascribed the prolonged clotting time of shock to a circulating antithrombin. Evidence that the anticoagulant in anaphylactic shock is heparin has been presented by Jacques (*7*) and Jorpes *et al.* (*8*), who hold that it is released from the mast cells. More recently, Dougherty and Dougherty (*3*) have shown similar changes in the mast cells of the experimental animal receiving 11-dehydro-17-hydroxycorticosterone (cortisone). Selye (*12*) has stated that the "alarm reaction," in general, decreases the clotting time and has postulated that the frequent occurrence of thrombosis following injuries or surgical procedures may be related to humoral factors released in response to stress. In man, disease with inflammation, necrosis, and thrombosis have each been shown by Schilling and DeNatale (*11*) to be accompanied by significant changes in the prothrombin times, which are not necessarily reflections of altered heparin levels.

In the light of these observations and on the premise that exogenous adrenocorticotropic hormone (ACTH) induces some changes in the human being that qualitatively simulate the response to stress, it appeared desirable to determine what effect this hormone and cortisone might have on the coagulation mechanism. To date, observations during hormonal therapy have been made on 20 patients receiving ACTH[1] and on 6 receiving cortisone[2] for the experimental treatment of ocular and certain collagen diseases. In addition, selected control studies have been made on 3 patients with Addison's disease, on 1 with hypopituitarism, on 1 individual with fatigue and diarrhea, and on 3 normal persons.

[1] Obtained from the Armour Laboratories, Chicago, through the cooperation of John R. Mote, medical director.

[2] Obtained from the Merck Laboratories, Rahway, N. J., through the courtesy of J. M. Carlisle, medical director.

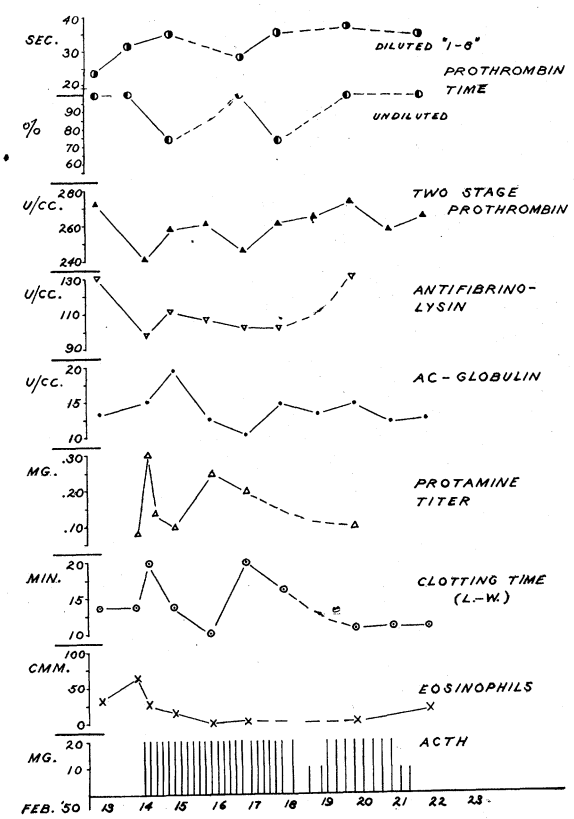

FIG. 1.

The following blood studies were made: clotting times by modified Lee-White and/or Howell procedures; direct platelet counts using the Rees-Ecker solution; undiluted prothrombin times[3] according to Quick (*10*); "1 to 8" plasma-diluted prothrombin times[3] by the Link-Shapiro method (*13*); heparin or heparinlike levels by a modification of the protamine titration of Allen (*1*); direct eosinophil counts by a modified Forsham-Thorn technique (*5*); and plasma accelerator (ac) globulin, antifibrinolysin, and two-stage prothrombin levels, as determined in the laboratory of W. Seegers,[4] of Wayne University College of Medicine, Detroit.

In the 3 normal persons used as controls, a single 20-mg intramuscular injection of ACTH produced in 4 hr a significant increase in circulating heparin or heparinlike material, a parallel prolongation of the clotting time,

[3] We are indebted to F. W. Hartman and V. Schelling, of the Division of Laboratories, Henry Ford Hospital, for these determinations.

[4] We express our thanks to Dr. Seegers for his generous help in these studies.

and the usual reduction in the circulating eosinophils.

These changes are represented on the left side, lower portion of Fig. 1, following the initial 20-mg injection of ACTH in this normal male. In the 2 normals receiving only one dose of ACTH, a return to the control levels was noted by the 8th hr. In the one (Fig. 1) who continued to receive 20 mg every 4 hr, the protamine titer and clotting time had reached control levels by the 20th hr, and the eosinophil count continued the expected decline.

The effects of ACTH and cortisone on blood clotting time and on certain of the contituents known to be active in the coagulation mechanism are depicted graphically in Figs. 1, 2, 3, and 4, which have been selected from the series. They are not representative of the group as a whole but illustrate, rather, the scope of the induced changes, as well as the differences that have been observed. In general, there are no consistent changes from patient to patient. The alterations following these hormonal agents appear to be, in part, a function of both the initial level of adrenal cortical activity and the integrity of the coagulation mechanism itself, which existed prior to the hormonal administration. This belief is supported, in part, by our observations on the patients with Addison's disease, with hypopituitarism, and on the individual with fatigue and diarrhea. All these persons had the expected high eosinophil counts of adrenal insufficiency and in all, the clotting times and protamine titers were above our upper limit of normal (0.10–0.14 mg for the latter). Abnormally high plasma-diluted

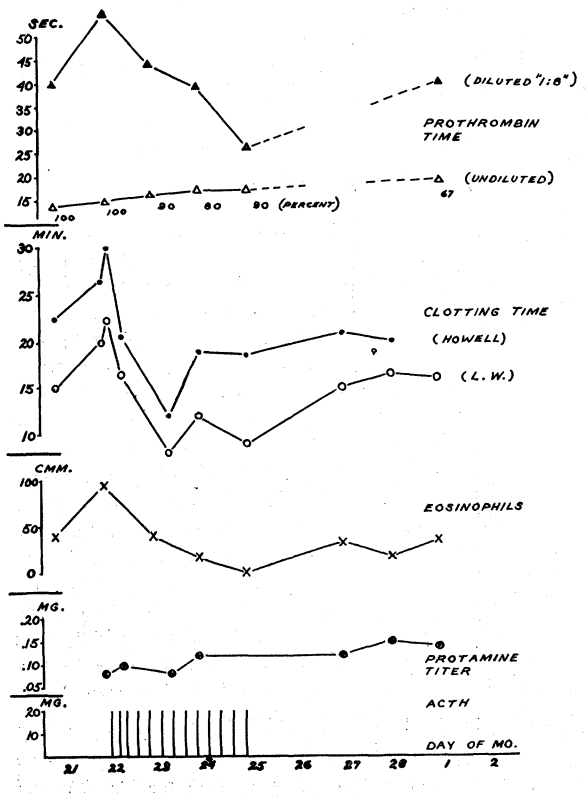

FIG. 2.

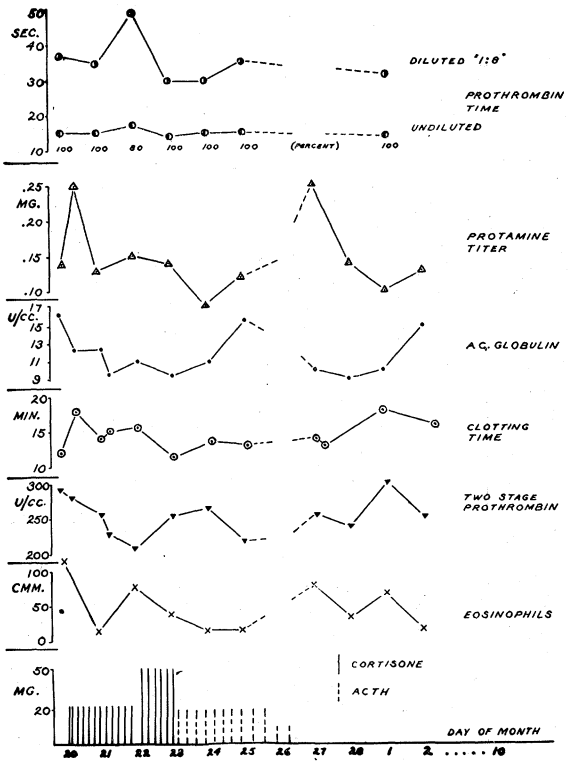

FIG. 3.

prothrombin times were observed in one Addisonian, whereas in 2 others the values were normal.

Fig. 2 demonstrates the profound changes in coagulation time that may follow ACTH administration. The clotting time by the two methods, in less than 24 hr, was reduced by 15 min. In this patient the reduction was paralleled by significant changes in the prothrombin system, as reflected in the marked shortening of the plasma-diluted prothrombin times. It is notable that the control level of circulating heparin or heparinlike material was so low as to suggest, and only suggest, a possible compensatory reduction for the initial prothrombin defect. In this individual, no significant increase in circulating heparin was observed at any time. The initial delayed diluted prothrombin time was not uncommonly encountered in this series. Of 20 patients adequately studied, 6 had the initial defect, and in every instance the diluted prothrombin time was brought to normal while ACTH or cortisone was being given.

In Fig. 4 are illustrated the changes that might be considered the most representatives of the series and that, in some respects, are similar to the alterations induced in the one normal male (Fig. 1) under continued ACTH administration. The following can be noted to have occurred: initial transient increase in protamine titers and clotting times; slight but significant lowering of clotting times during therapy and return to control level when the hormone was withdrawn; a varying increase in the level of circulating heparin or heparinlike material on therapy which, inexplicably, continued after ACTH withdrawal; significant changes both in the percent prothrombin and

the diluted prothrombin times, which appear inverse to the changes in protamine titers. These return to normal on completion of therapy and can be correlated with the level of adrenal cortical activity by the number of circulating eosinophils (plotted in the lower portion of the figure). Of 20 patients studied, following the first 24 hr of treatment, 12 had an over-all decrease, 6 had no significant change, one had an increase in clotting time, and one had insufficient observations to interpret adequately. Seven of 17 patients with normal initial values had a significant increase in the diluted prothrombin time on therapy. Two had lowered and 5 had elevated protamine

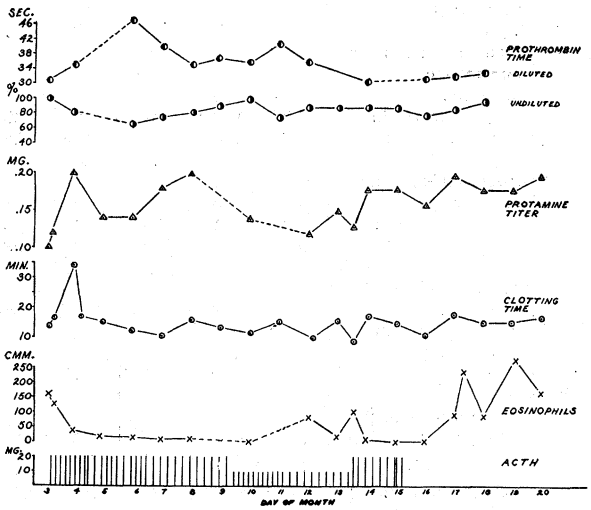

FIG. 4.

titers, of 17 patients in whom this was adequately studied during the period of hormone administration. In addition, there were several patients with high initial protamine titers and low initial eosinophil counts suggesting pre-existing stress, and in these significant changes on treatment were not observed. Platelet counts were not significantly altered.

Figs. 1 and 3 are presented to illustrate the influence of ACTH and cortisone on a number of humoral factors not covered above and known to be active in the coagulation process. It is apparent that variations were produced in the levels of plasma ac-globulin and two-stage prothrombin (Fig. 3) and in ac-globulin and antifibrinolysin (Fig. 1). At present the significance of these changes to blood coagulation in patients receiving ACTH or cortisone is not apparent and will not be further discussed. Similar observations have been made on 6 additional patients under hormonal treatment.

The results of the study herein reported do not allow any broad conclusions, inasmuch as the changes following ACTH and cortisone are by no means uniform from patient to patient. There is little question, however, that the adrenal cortex exerts an effect on a number of constituents of the clotting mechanism and, in some instances, may alter significantly the clotting time when its function is accelerated. The release of heparin or a heparinlike substance into the blood following ACTH or cortisone may be akin to the hyperheparinemia of anaphylactic shock. It affords an additional link in our present understanding of the interrelationship of the mast cell with its heparin production, the adrenal cortex, and the changes in blood coagulability which may accompany the response to stress. The application of the results of the present study to the patient with adrenal insufficiency and to the patient undergoing surgery, among many problems, is obviously indicated and is being currently pursued.

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The Nucleus-Dependence of P^{32} Uptake by the Cell

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The various lines of evidence concerning the role of the nucleus in living cells converge in the hypothesis of an ill-defined "determination," or "control," function. The most complete evidence, that from studies of reproduction, heredity, and morphogenesis, is based on experimental designs that give little insight into the problem of the function of the nucleus in the current activities of the mature cell. The fragmentary information on this latter problem, derived from observations on cells deprived of nuclei, has generally been interpreted in terms of two hypotheses (*1*): (1) that the nucleus is a center of essential energetic processes and is immediately involved in the short-term metabolism of the cell, or (2) that the nucleus is concerned only with the long-term maintenance of the cytoplasm. The latter hypothesis derives from observations on cells whose nuclei have been removed. These may survive for some time (a few days to several months [*4*]) in terms of most identifiable activities, but ultimately they decline and die.

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[2] Atomic Energy Commission Postdoctoral Fellow, 1949–50.