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THE ROLES OF CORTISONE, DESOXYCORTICOSTERONE, AND ADRENALINE IN PROTECTING ADRENALECTOMIZED ANIMALS AGAINST HAEMORRHAGIC, TRAUMATIC, AND HISTAMINIC SHOCK

BY

B. N. HALPERN*, B. BENACERRAF, AND M. BRIOT

From the Laboratory of Experimental Medicine of the Clinique Médicale Propédeutique de l'Hôpital Broussais (Prof. Pasteur Vallery-Radot) and Centre National de la Recherche Scientifique

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The secretions of the adrenal cortex play an undisputed role in the defence of the organism against all sorts of noxious agents (Swingle and Remington, 1944). Adrenalectomy renders an animal much more susceptible to a variety of traumatic procedures, as well as to the injection of endogenous or exogenous toxins, all factors which in the end have a damaging effect upon the peripheral vascular system. The adrenalectomized animal, when submitted to these noxious agents, eventually dies in circulatory failure, which can be attributed to a collapse of the peripheral circulation (Wyman and Tum Suden, 1939 ; Selye, 1949 ; Halpern and Wood, 1950a, b).

Treatment with whole cortical extracts or with desoxycorticosterone (Swingle and Remington, 1944) restores to the adrenalectomized animals at least part of their natural resistance to various forms of stress. Many hypotheses have been advanced to explain the mode of action of these hormones in protecting animals against stressing agents, but as yet no definite evidence substantiating these hypotheses has been reported. Moreover, it is not clear whether any one hormone is primarily responsible for this protection, or whether desoxycorticosterone as well as the glucocorticoids are equally able to restore to the adrenalectomized animal its natural resistance. These considerations led us to investigate whether cortisone exhibits a similar protective action against various forms of vascular shock such as haemorrhagic, traumatic, or histaminic shock, and whether the protective actions of cortisone and of desoxycorticosterone are comparable. A study of the mechanism by which the protection of these hormones is effected has been attempted.

HAEMORRHAGIC SHOCK

METHODS

The experiments were performed on white male rats weighing 125±25 g. The adrenal glands were removed in one stage by the dorsal route under light ether anaesthesia. Immediately after the operation, the animals received 1.5 mg. desoxycorti-

* Address: Hôpital Broussais, 96, Ruc Didot, Paris (14) France.

costerone, and 2 ml. isotonic saline subcutaneously. They were allowed access to a normal diet with plenty of fluids. Mortality due to the operation was small (less than 10 per cent).

The haemorrhages were carried out three days after the adrenalectomy. Repeated bleedings of 0.5 ml. blood (with a maximum number of nine each) were carried out every five minutes by puncture of the ophthalmic venous plexus with a calibrated glass pipette through the inner angle of the eye (Halpern and Pacaud, 1951). The number of bleedings performed was proportional to the weight of the animals and was calculated so that the total amount of blood taken was equal to about 50 per cent of the theoretical blood volume. A number of control animals died before this amount could be withdrawn. Into all the animals who survived this procedure, the total amount of blood which had been withdrawn and heparinized was slowly reinjected intravenously. The animals were observed in the hours and days that followed and the number of deaths recorded. The animals treated with cortisone acetate or desoxycorticosterone received 5 mg. of drug the day before they were bled and again four hours before bleeding.

The rectal temperature was taken and recorded regularly throughout the experiment. The haemoglobin concentration of the blood was measured at intervals during the haemorrhage in order to ascertain the shift of fluids from the extracellular to the vascular compartment.

In order to investigate the effect of the bleedings on the arterial blood pressure, this was recorded in the carotid artery in a number of animals. The animals were anaesthetized lightly with ether or with urethane (100 mg. per 100 g.) and a small cannula introduced into the carotid artery. The haemorrhages were then carried out as described above.

Results

Control animals.—The results of these experiments are presented in Fig. 1. Out of ten control animals, only one survived for a few hours the full bleeding, which corresponded to 50 per cent of its theoretical blood volume, in spite of being transfused with the total volume of blood removed. The nine other animals died in the course of the bleedings, starting after the fourth bleeding.

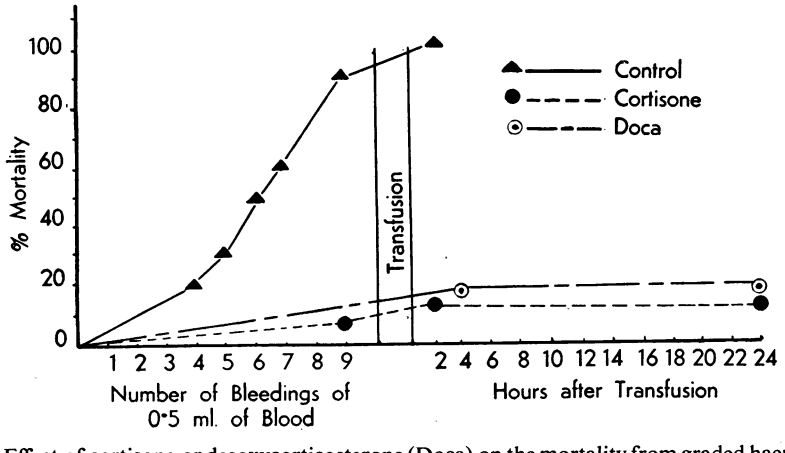

FIG. 1.—Effect of cortisone or desoxycorticosterone (Doca) on the mortality from graded haemorrhage involving 50 per cent of their blood volume in adrenalectomized rats.

All the control animals exhibited a progressive drop in rectal temperature during the bleedings as shown in Fig. 2; when an animal survived all the bleedings, its temperature fell to 30° C.; obviously, when a rat died in the early stages of the experiment, the fall of temperature was less striking.

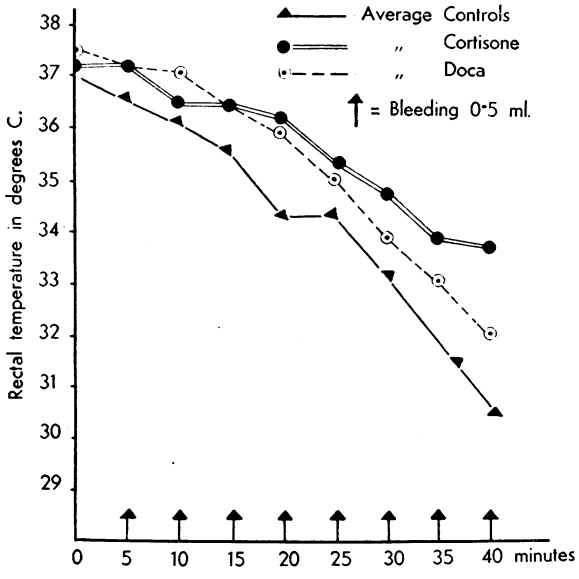

FIG. 2.—Effect of cortisone or desoxycorticosterone (Doca) on the rectal temperature of adrenalectomized rats when they sustain a graded haemorrhage involving 50 per cent of their blood volume.

Measurements of the haemoglobin concentration in the blood during the experiment are presented in Fig. 3. They indicate that in the early stages there is a slight haemoconcentration, corresponding presumably to the call on blood reserves, but in later stages there is a slight haemodilution, caused probably by absorption of fluid from the extracellular compartment.

Two typical records of the blood pressure after each bleeding are presented in Fig. 4. Generally, the successive bleedings produce a fall of blood pressure, which is relatively sharp at the time of the bleeding. The fall of blood pressure between two successive bleedings is usually 1 to 2 cm. Hg. Usually, after the seventh or eighth bleeding, the animals die suddenly owing to vascular collapse. Two animals maintained a relatively high blood pressure until the fifth bleeding, but it then fell progressively and severely. Two animals, who survived nine bleedings and were transfused with their own blood, showed a return of blood pressure to a nearly normal value, but could not maintain it and, after a fresh fall of blood pressure, died within an hour.

Animals treated with cortisone.—Out of fourteen treated animals, none died before the total amount of blood had been withdrawn; one died after the ninth bleeding, and another one died two hours after being transfused; twelve animals were alive after twenty-four hours, and may be considered as having definitely recovered (Fig. 1).

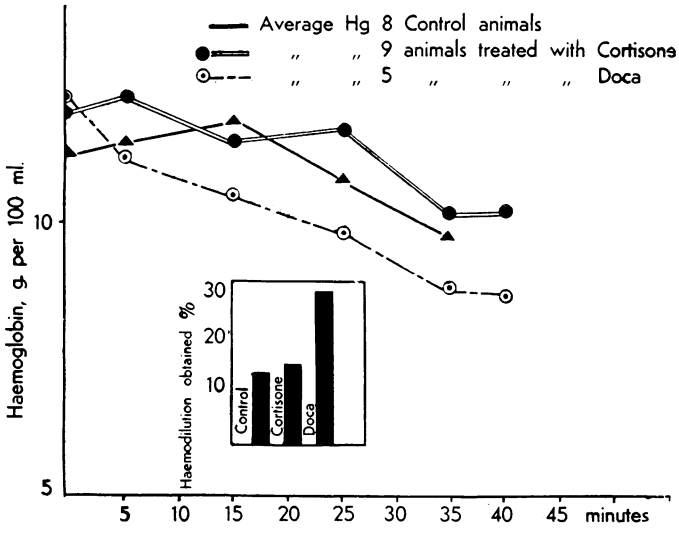

FIG. 3.—Effect of cortisone or desoxycorticosterone on haemoglobin concentration of the blood and haemodilution in adrenalectomized rats while they sustain a graded haemorrhage involving 50 per cent of their blood volume.

It must be stressed (see Fig. 2) that animals treated with cortisone tend to maintain their body temperature much more efficiently than the controls. A study of the haemoconcentration curves (Fig. 3) reveals no significant difference between cortisone-treated animals and the controls. However, analysis of the blood-pressure records (Fig. 4) shows a strikingly different behaviour between cortisone-treated and control animals. The blood pressure of the treated rats is generally higher by an average of 1.5 cm. Hg than that of the controls. During the haemorrhages the fall of blood pressure in these animals seldom becomes significant before the fifth bleeding. After this, when it starts to drop, the slope of the blood pressure is much less steep than in the control animals. Arterial pressure never falls to very low levels, and after transfusion the blood pressure rises and is maintained at a normal level.

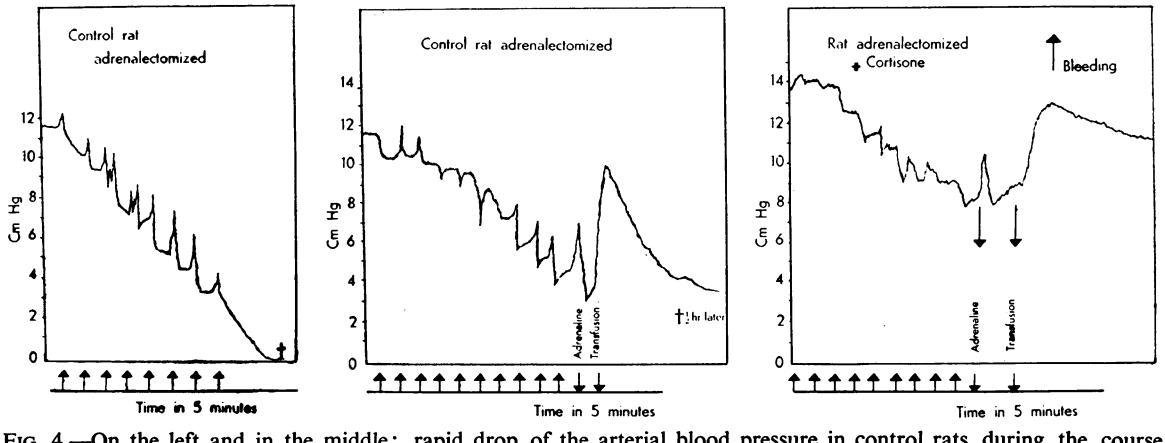

FIG. 4.—On the left and in the middle: rapid drop of the arterial blood pressure in control rats during the course of the bleedings. On the right: record of blood pressure in a cortisone-treated rat who sustained the same haemorrhagic procedure and was transfused after the ninth bleeding.

Animals treated with desoxycorticosterone.—Out of six treated animals, one died a few hours after being transfused, and the remainder survived twenty-four hours and may be considered as having definitely recovered. The rectal temperatures of these animals were, on the average, higher than those recorded for the control group, but lower than those of the cortisone-treated animals (Fig. 2).

Study of the haemoconcentration (Fig. 3) reveals a significant difference between the rats treated with desoxycorticosterone and those treated with cortisone and the controls. Even in the early stages there is no haemoconcentration, but rather a progressive haemodilution which in the end reaches an average of 28 per cent of the original haemoglobin concentration; in the control and the cortisone-treated animals, on the other hand, it reached an average value of only 14 per cent. There is, in these animals, a greater transfer of fluids from the extracellular spaces into the vascular bed, which much improves the chances of recovery. The blood pressures, in this group, during the haemorrhages were not very different from those of the control rats, except that, after transfusion of the withdrawn blood, the animals treated with desoxycorticosterone maintained their normal blood pressure much better than the control animals, but not quite as well as those treated with cortisone.

Limb Ischaemia Shock

Methods

These experiments were performed on 51 rats three days after adrenalectomy; the rats were adrenalectomized and treated as described above. The animals treated with cortisone acetate or with desoxycorticosterone received subcutaneously 5 mg. of the drug the day before the application of the tourniquet. Another 5 mg. was administered two hours before application of the garrotte, and 1.5 mg. six hours after the removal of the tourniquet.

Rubber tourniquets were applied at the origin of the thigh of the left leg in one group of animals, and to the same site on both legs in another group; they were maintained for two hours, the animals being kept in separate cages. The tourniquets were then removed and the limbs massaged gently. The animals were kept in a well-heated room. Haemoconcentration was measured from the change in haemoglobin concentration.

Results

The results of these experiments are presented in Table I. It can be seen that, when the tourniquet was applied to one limb, 7 out of 10 controls died in the 12

TABLE I

EFFECT OF CORTISONE AND OF DESOXYCORTICOSTERONE ON MORTALITY FROM LIMB ISCHAEMIA IN ADRENALECTOMIZED RATS

Limb ischaemia One limb—2 hours Mortality at 24 hours			Limb ischaemia Two limbs—2 hours Mortality at 24 hours		
Control	After cortisone	After desoxycorti-costerone	Control	After cortisone	After desoxycorti-costerone
7/10	0/11	0/6	11/12	0/10	0/7

hours that followed the removal of the tourniquet. All the animals treated with cortisone or with desoxycorticosterone survived. When the tourniquets were applied to two limbs, 11 out of 12 control animals died, and again all the animals treated with cortisone or desoxycorticosterone survived.

The changes in haemoconcentration in the control animals and in those treated with cortisone when the tourniquet was applied to one limb were studied. No significant difference between the control and the cortisone-treated animals was apparent. It should be noted that the maximum haemoconcentration observed was relatively small, and far from sufficient to explain the death of the control animals on the basis of loss of fluid in the limb.

Histaminic Shock

It is well known that mice are relatively insensitive to histamine. According to Halpern and Wood (1950a, b), the lethal dose of histamine dihydrochloride, injected intraperitoneally, is 50 mg. per 20 g. of body weight for normal mice. Adrenalectomy renders mice considerably more sensitive to histamine, so that it requires only 0.5 mg. per 20 g. of histamine to kill an adrenalectomized animal (Halpern and Wood, 1950b). It has been shown by the same authors that adrenaline increases the resistance of the adrenalectomized mice to histamine, but only raises the lethal dose of histamine from 0.5 mg. to 5 mg. per 20 g. Adrenaline therefore does not seem to be the only factor responsible for the high resistance of normal mice to histamine.

Methods

One hundred and twenty-four adult mice (female) weighing about 20 ± 5 g. were used for these experiments. Bilateral adrenalectomy was performed under light ether anaesthesia and the animals were allowed access to a normal diet with plenty of fluid. Immediately after the operation they received 0.5 mg. desoxycorticosterone and 0.5 ml. isotonic saline subcutaneously. The animals treated with cortisone acetate or desoxycorticosterone were injected with 0.5 mg. of drug twice a day for three doses, the last dose being given 4 to 5 hours before the injection of histamine. Histamine dihydrochloride was administered intraperitoneally in 0.5 ml. isotonic saline per 20 g. of body weight 48 hours after the operation. Two groups of animals treated with cortisone or desoxycorticosterone were also treated with adrenaline. To these animals and to a number of controls, 20 μg. adrenaline per 20 g. body weight was injected subcutaneously 30 min. before the injection of histamine. In some animals, haemoconcentration was measured by the change in the haemoglobin concentration of the blood. As in the rat, blood samples were obtained by puncture of the ophthalmic venous plexus through the inner angle of the eye with a capillary glass pipette (Halpern and Pacaud, 1951).

Results

The results of these experiments are presented in Table II and in Fig. 5. They show that cortisone can increase the tolerance of adrenalectomized mice to histamine from 0.5 mg. to 2.5 mg. per 20 g., whereas desoxycorticosterone is not capable of protecting the animals against one lethal dose of histamine. This protective power of cortisone, however, does not restore to the animals their ability to withstand doses of histamine tolerated by normal mice. In contrast to this weak protective power of cortisone alone, treatment with both adrenaline and cortisone

TABLE II

EFFECT OF CORTISONE, DESOXYCORTICOSTERONE, AND ADRENALINE ON HISTAMINE TOXICITY IN ADRENALECTOMIZED MICE

Histamine mg./20 g.	Control	After adrenaline	After cortisone	After desoxycorticosterone	After desoxycorticosterone and adrenaline	After cortisone and adrenaline
0.25	3/9			1/3		
0.5	10/10		0/10	4/6		
2.5			5/14	4/4		
5.0		1/3	10/15			
10.0		4/5			2/8	0/6
17.5					2/5	
25.0					4/5	2/15
50.0						7/7

confers on adrenalectomized mice the ability to withstand the same doses of histamine as normal mice. It should also be noted that, although desoxycorticosterone has practically no protective power by itself, since it cannot protect against one lethal dose of histamine, it is able to increase considerably the antihistaminic power of adrenaline in the adrenalectomized mouse, although to a much lesser degree than cortisone.

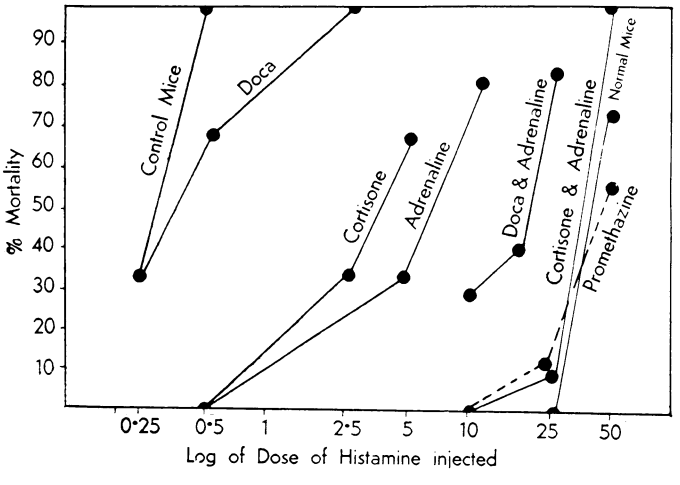

FIG. 5.—Effect of cortisone, desoxycorticosterone (Doca), and adrenaline on the toxicity of histamine in adrenalectomized mice. The data concerning promethazine (dotted line) have been extracted for comparative purposes from the study by Halpern and Wood (1950b).

The degree of tolerance to histamine afforded by cortisone and adrenaline is reflected in the degree of haemoconcentration (Table III). When the animals are protected with cortisone, or cortisone and adrenaline, the degree of haemoconcentration which usually results from the injection of histamine diminishes. Thus the degree of haemoconcentration observed in adrenalectomized control animals, who received 0.5 mg. per 20 g. histamine, was 65 per cent. In animals treated with cortisone, the same dose of histamine produced an increase of haemoconcentration of only 12 per cent ; when adrenalectomized mice were treated with cortisone, a

TABLE III

EFFECTS OF CORTISONE AND ADRENALINE ON HAEMOCONCENTRATION PRODUCED BY HISTAMINE IN ADRENALECTOMIZED MICE

Second blood sample taken 20–30 min. after histamine injection. Haemoconcentration measured from change in haemoglobin concentration.

Treatment	Average haemoconcentration per cent after histamine 2 HCl (mg.)					
	0.25	0.5	2.5	5.0	10	25 mg.
Control mice	31	65				
After 20 μg. adrenaline HCl ..				25	47	
After cortisone		12	31	50		
After cortisone + 20 μg. adrenaline HCl ..					2	8

dose of histamine as high as 2.5 mg. per 20 g. brought about an increase of haemoconcentration of 31 per cent. Moreover, when the animals were treated with both cortisone and adrenaline, they tolerated as much as 25 mg. histamine per 20 g., and the average haemoconcentration was only 8 per cent.

Discussion

The results reported above indicate that cortisone provides the adrenalectomized animal with a definite protection against haemorrhagic shock. Whereas control animals die early with symptoms of vascular collapse, treatment with cortisone makes it possible for adrenalectomized animals to survive repeated bleeding and to maintain relatively high blood pressures and body temperatures during the procedure. This protective effect is probably due to an improvement of the vascular tone of the small capillaries and arterioles, the animals being able to maintain, in spite of drastic reductions of blood volume, arterial blood pressures sufficient for the safeguarding of essential life processes. It is well known that the arterioles of adrenalectomized animals are not able to compensate for loss of blood and show a decreased sympathetic tone (Swingle and Remington, 1944 ; Wyman and Tum Suden, 1939) ; considerable evidence, both direct and indirect, indicates that the capillaries of the adrenalectomized animal in insufficiency are atonic, dilated, and abnormally permeable (Wyman and Tum Suden, 1939 ; Zweifach and Chambers, 1942 ; Selye, 1949).

Treatment with cortisone restores normal function to these vessels in adrenalectomized animals. It should be noted that the animals treated with cortisone generally exhibit a higher blood pressure than the control animals, and this can also be interpreted as being due to an increase of vascular tone.

Treatment with desoxycorticosterone also protects the animal against death from haemorrhagic shock, but the effects on the blood pressure, the temperature, and the changes in haemoconcentration are somewhat different than those seen in animals treated with cortisone. Desoxycorticosterone does not maintain either blood pressure or temperature as well as cortisone does. The greater haemodilution observed in animals treated with desoxycorticosterone at the end of the bleeding could be a considerable contributing factor to survival. Desoxycorticosterone causes a water

and salt retention in the extracellular fluids, with easier transfer of fluids into the vascular compartment when the need arises.

The results obtained in the limb ischaemia experiments show that cortisone and desoxycorticosterone protect the animal equally well against death in the crush syndrome. Death in limb ischaemia is probably due to vascular collapse resulting from absorption of angiotoxic products from the ischaemic limb. The relatively small degree of haemoconcentration in these animals and the lack of difference between the haemoconcentrations of the treated and the control groups, in spite of the difference in mortality, indicate that the loss of plasma fluid is not the most important factor in the mechanism of the vascular collapse.

The absorption of toxins from the ischaemic limb can cause a massive peripheral vasodilatation, with decreased blood pressure, stasis, and vascular collapse. It is probable that the cortical hormones act here on the vascular tone, through the same mechanism as in haemorrhagic shock, without it being necessary to invoke a specific detoxifying mechanism to explain the action of the hormones. These facts should be connected with the observation that the adrenalectomized animal shows a much poorer ability to perform muscular work than the normal animal, in spite of the fact that the isolated muscles from both animals perform equally well (Ramey, Goldstein, and Levine, 1950). This decreased ability to perform muscular work has been ascribed to the asthenia of the blood vessels and the poor irrigation of the organ at work. It can be restored to normal by treatment with cortisone.

The results obtained in histaminic shock indicate that cortisone is much more active than desoxycorticosterone in its capacity to confer on the adrenalectomized animal resistance to vascular injuries.

The animals treated with cortisone can tolerate a dose of histamine about five times larger than the control adrenalectomized animals. In this respect, cortisone behaves much like adrenaline alone in these adrenalectomized animals. None of these substances alone can restore the normal tolerance to a single injection of histamine, as does promethazine (Halpern and Wood, 1950b). However, treatment with adrenaline of adrenalectomized mice, which have been receiving cortisone for 24 hours, confers on them the ability to withstand the high doses of histamine tolerated by the normal mice.

Although treatment of animals receiving desoxycorticosterone with adrenaline does not bring back the tolerance to histamine to the normal level, as it does for cortisone-treated animals, it makes it possible for the animal to withstand higher doses of histamine than does treatment with adrenaline alone.

These observations suggest that cortisone, and to a lesser extent desoxycorticosterone, maintain the tone of blood vessels by improving the ability of the vascular bed to respond to vasoconstrictor agents. It must be mentioned that Levine and his co-workers (Ramey, Goldstein, and Levine, 1951 ; Fritz and Levine, 1951) have already observed that treatment of adrenalectomized animals with cortisone increases and regulates the vascular effect of noradrenaline. This interpretation of the mode of action of cortisone on small vessels is in harmony with the numerous reports that the sensitivity to injected rennin is lost in adrenalectomized animals, and is restored only by cortical extracts (Friedman, Somkin, and Oppenheimer, 1940 ; Remington, Collings, Hays, Parkins, and Swingle, 1941). It is also consistent with the observation of Selye (1950) that adrenaline promotes only a temporary rise

in the resting blood pressure of adrenalectomized animals, and that their tendency to develop severe hypotension during stress cannot be effectively combated by this hormone. The vasoconstrictor effect of adrenaline in the mesentery of the rat similarly declines progressively after adrenalectomy.

The findings reported here indicate that cortisone plays a predominant role in the resistance of animals to traumatic and toxic injuries: cortisone seems to enhance this resistance by improving the compensatory vascular reactions which are markedly depressed during the state of shock. The results obtained in experiments involving histaminic shock suggest that cortisone, and to a lesser degree desoxycorticosterone, are potentiating the effect of adrenaline in combating the angiotoxic effects of histamine.

SUMMARY

1. The actions of cortisone, desoxycorticosterone and of adrenaline on haemorrhagic, traumatic, and histaminic shock in recently adrenalectomized animals have been investigated.

2. In haemorrhagic shock induced in adrenalectomized rats by repeated and graded haemorrhage, cortisone, and to a lesser extent desoxycorticosterone, each produce a definite protective effect. Whereas 90 per cent of control animals die early with symptoms of irreversible vascular collapse, treatment with cortisone makes it possible for animals subjected to bleeding to survive and to maintain a relatively high blood pressure and body temperature. Desoxycorticosterone cannot maintain either the blood pressure or the body temperature so well as cortisone does.

3. In animals submitted to limb ischaemia either cortisone or desoxycorticosterone protect the animals equally well against death from crush syndrome. Animals dying from limb ischaemia die probably from vascular collapse owing to the absorption of angiotoxic products from the ischaemic tissue. The relatively small haemoconcentration found in these animals and the lack of difference in the degree of haemoconcentration between the treated and the control animals, in spite of the drastic difference in mortality, indicate that the loss of the plasma fluid is not the most important factor in the mechanism of the vascular collapse.

4. The results obtained in histaminic shock indicate that desoxycorticosterone does not modify the high sensitivity of adrenalectomized mice to histamine, whereas cortisone increases this tolerance about five times and adrenaline about ten times. None of these three hormones alone can restore the natural resistance to histamine in the adrenalectomized animal. However, in adrenalectomized mice receiving cortisone, treatment with adrenaline confers the ability to tolerate doses of histamine as great as those tolerated by the normal animal. Desoxycorticosterone associated with adrenaline has a similar effect although it does not restore the tolerance to the normal level.

5. The results indicate that cortisone and, to a lesser degree, desoxycorticosterone enhance the resistance of animals to traumatic and toxic injuries by improving the ability of the small vessels to respond to the exogenous and endogenous vasoactive agents.

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