SOME EFFECTS OF PITUITARY ADRENOTROPIC HORMONE (PATH), EXTRACT OF SUPRARENAL CORTEX, AND COLCHICINE ON THE HAEMOPOIETIC SYSTEM

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Only scant information is available concerning the mechanism whereby the level of the blood cells is controlled, and a balance maintained between new cells entering the blood and old cells leaving it, or being destroyed while in the circulation. The lymphocytes would appear to lend themselves best to the investigation of such a mechanism, for we can in their case obtain an approximate idea as to both the numbers entering and the numbers leaving the blood.

The view that the blood cells are subject to hormonal rather than direct nervous control has long been favoured by many haematologists, partly because of the poor innervation of haemopoietic tissues, and partly because both in the blood and the parent tissues the cells are subject to continual displacement. In the case of lymphocytes and lymphoid tissue, a number of observations have in the past seemed to point to the suprarenals as an essential part of the control mechanism (Drinker & Yoffey, 1940). Recently, attention has been focused still more closely on the suprarenals since the discovery and investigation of pituitary adrenotropic hormone, or for short PATH.

On the question of terminology, a number of workers designate the adrenotropic hormone of the pituitary as adrenocorticotropic hormone, or briefly ACTH. This appears to be a somewhat loose designation, for other hormones are known to have an action on the cortex. Thus Vogt (1944) has shown that adrenaline stimulates cortical secretion. Further, the thyroid gland also has important effects on the suprarenal cortex (Hoskins, 1910; Ingle & Higgins, 1938; Swann, 1940). It may be that in many animals this is effected through the pituitary, but certainly not in all. In the pigeon, Miller & Riddle (1939) have shown that thyroxine induces cortical hypertrophy even in the hypophysectomized animal. Unpublished results by one of us (J.S.B.) indicate further that the action of the pituitary adrenotropic hormone, while it may be primarily on the cortex of the suprarenal, produces changes in the medulla also. For this reason the term pituitary adrenotropic hormone (PATH), indicating the specific origin of the hormone in question, as well as its possible action on the suprarenal as a whole, seems preferable to ACTH, and is accordingly employed in this paper.

Dougherty & White (1944) found that 'Single injections of pituitary adrenotropic hormone in

mice, rats and rabbits produce within a few hours an absolute lymphopenia...'. White & Dougherty (1945) further noted that daily injections of PATH, continued for 15 days, produced in the blood of mice (a) increase in haemoglobin and red cells, (b) an absolute lymphopenia, and (c) an increase in polymorphs; concurrently there was diminution in weight of lymph nodes and thymus (Dougherty & White, 1943). They also reported unpublished studies which indicated 'that in mice injected with adrenotropic hormone daily for a period as long as one year, a persistent absolute lymphopenia is present'. Simpson, Li, Reinhardt & Evans (1943) reported atrophic changes in lymph nodes and thymus after administration of PATH, and stated that in the lymph nodes a 'flushing out' of lymphocytes occurred. Ingle (1938) noted atrophy of the thymus following the administration of cortical extract, as also did Crede & Moon (1940) after PATH. Wells & Kendall (1940) described thymic involution after cortico-sterone. Reinhardt & Li (1945) observed a fall in the lymphocyte content of thoracic duct lymph in rats after PATH, and a similar observation has been made by Yoffey, Reiss & Baxter (1946) in cats.

Dougherty & White (1945) studied in detail the early histological changes in mouse and rabbit lymphoid tissues after the administration of PATH, and cortical extract.

The present experiments were undertaken to confirm the findings of White & Dougherty (1945) and to see whether the administration of PATH and extract of suprarenal cortex for a period of 1 month, as compared with 15 days in their published work, would give more clear-cut results.

MATERIAL AND TECHNIQUE

The experiments were performed on twenty-six healthy Wistar rats and two rabbits. Cortrophin (Organon)[†] was the preparation of PATH used, standardized in Sudanophobic units as described by Reiss, Balint, Oestreicher & Aronson (1936). The extract of suprarenal cortex employed in the rats was Eschatin,[‡] standardized in dog units (Harrop, Pfiffner, Weinstein & Swingle, 1932); in the rabbits another commercial extract, also

standardized in dog units, was employed. Rat blood was obtained from the tail vein, rabbit blood from the ear. Before incising a tail vein, rats were anaesthetized with ether, a procedure which Crafts (1944) and others have shown to be without significant effect on the blood count. For the routine staining of blood films Wright's stain was employed, and in the differential counts not less than 200 cells were counted. Reticulocyte staining was done with a dried film of brilliant cresyl blue, counting 1000 cells.

Sections of lymph nodes—mesenteric or cervical—and of thymus were cut at 6μ after fixation in Zenker-formol and paraffin embedding, and were stained with iron haematoxylin and Dominici's eosin-orange G-toluidin blue stain.

The experiments fall into four groups:

I. Twelve rats which were given daily intraperitoneal injections of PATH, cortical extract, or both (for dose see Table 1) for periods ranging from 3 to 21 days. In addition these rats were given between 0·1 and 0·2 mg. colchicine per 100 g. body weight 16–17 hr. before being killed. Three blood counts were made on each animal: count A immediately before beginning the injection of PATH or cortical extract, count B 3–5 hr. after the first injection, and count C just before the animal was killed. These experiments lasted for periods ranging from 3 to 21 days.

II. Eight rats were given daily intraperitoneal injections of PATH or cortical extract (for dose see Table 2) for 27–28 days, but no colchicine. Colchicine had been given to group I in order to study more accurately details of lymphocyte mitoses. At that time we were not aware that colchicine itself had a markedly destructive effect on lymphoid tissue in rats, exactly like that noted in mice by Lits, Kirschbaum & Strong (1938). Accordingly in the animals of group II colchicine was omitted, but the administration of PATH and cortical extract continued for 4 weeks. Two blood counts were made in this group: count A, immediately before beginning the injections, and count B before killing the animals.

III. Six colchicine controls. In three animals colchicine was injected and the animals were killed 16–17 hr. later, for histological examination of lymph nodes and thymus. In another three animals the blood picture in response to colchicine was also investigated. Count A was made, then colchicine was injected (0·15 mg./100 g. body weight), and 16 hr. later count B was made and the animal was killed.

IV. Two rabbits, in which the red and white blood cells were counted over a period, to get the range of normal fluctuations, and then a cortical extract (c. 12·5 dog units per kg. body weight) was given subcutaneously every day for 16 and 40 days respectively.

RESULTS

Blood. Table 1 gives the results of the first series of twelve rats. Three blood counts were made in each experiment. Count A is the normal count before the injection commenced, count B was taken at times varying from 3 to $5\frac{1}{4}$ hr. after the first injection, and count C after the injections had continued for times ranging from 3 to 21 days. In this group count C was complicated by the administration of colchicine about 16 hr. before it was made, but count B shows the uncomplicated action on the blood lymphocytes of PATH and cortical extract, either separately or in combination. In eight out of the twelve experiments there was a marked fall in the blood lymphocytes; in one there was an appreciable rise, and in two no significant change; in one experiment count B was not recorded. On the whole the short term effect of PATH and of cortical extract in rats seems to be a fall in the blood lymphocytes, as noted by Dougherty & White (1944).

Table 2 presents the results of the second series of eight long-term rat experiments in which PATH and cortical extract were given over a 4-week period. PATH alone has induced a rise in the blood lymphocytes, but cortical extract alone or together with PATH a fall. In the case of the red cells also (Table 2) the results suggest the possibility that the action of PATH may differ from that of cortical extract, though the number of experiments is not big enough to draw definite conclusions.

In the twelve experiments quoted in Table 1, the long-term results are difficult to assess, for they are in part due to the colchicine. Eight experiments showed a fall in the blood lymphocytes, four a rise. In the case of the red cells four showed a fall and eight a rise, and in some the rise suggests an erythraemia. Furthermore, immature red cells (normoblasts) and white cells (a few myeloblasts, but chiefly myelocytes and metamyelocytes) appeared in the blood, sometimes in considerable numbers. At the same time marked basophilia was noted, though reticulocytes were not counted in this group. The appearance of immature red and white cells in rat's blood following the administration of colchicine, as well as a marked polymorphonuclear leucocytosis, agree with the findings of Dixon & Malden (1908), and later of the Brussels School (for further details see Lits *et al.* 1938).

The result in one rabbit experiment is presented in Text-fig. 1, which shows the daily blood lymphocyte counts during an experiment lasting 16 days, preceded by a control period of 22 days. The blood lymphocytes show a marked drop from the third to the eighth day of the experiment and then rise again. But the drop is of the same order as had occurred previously during the control period, and in any case was only temporary. In the second experiment, where injections were given

Table 1. *Blood lymphocytes per cu.mm. (ordinary type) and red blood cells in 1000's per cu.mm. (black type) after daily intraperitoneal injection into rats of Cortrophin* (C), *Eschatin* (E) *or Cortrophin + Eschatin* (C + E)

Count A immediately before injections commenced.
Count B 2¾–5¼ hr. after first injection.
Count C* after repeated injections for 3–21 days.

No. of animal	Count A	Count B	Count C	Time between A and B (hr.)	Time between B and C (days)	Colchicine mg./100 g. body weight	Substance used and daily dose per 100 g. body weight
29	19,920	25,036	14,425	5	8	0·15	C—0·14 s.u.
	—	—	—				
30	19,190	11,270	7,035	5¼	7	0·2	E—0·2 d.u.
	8,720	8,320	6,800				
31	12,670	13,254	7,870	4½	7	0·2	E—0·19 d.u.
	7,580	6,960	6,900				
32	16,826	7,090	10,937	4¾	17	0·175	C+E—0·2 s.u.+0·2 d.u.
	7,760	7,020	7,740				
33	13,532	—	8,487	—	18	0·2	C+E—0·4 s.u.+0·2 d.u.
	7,160	clotting	12,720				
34	14,998	11,240	13,920	4	18	0·18	C—0·4 s.u.
	8,620	7,680	7,760				
35	5,022	5,010	9,835	4¼	20	0·2	E—0·5 d.u.
	6,300	5,920	8,380				
36	13,993	8,597	17,036	4	4	0·2	C—0·25 s.u.
	8,420	7,810	9,780				
37	11,652	5,938	8,042	3	21	0·15	C—0·11 s.u.
	6,880	7,260	9,340				
38	8,050	3,310	5,040	5¼	21	0·1	C—0·22 s.u.
	7,020	7,780	9,740				
39	14,283	7,333	27,277	2¾	21	0·1	E—0·4 d.u.
	7,440	7,640	5,260				
40	9,045	4,673	15,379	3¾	3	0·1	C—0·67 s.u.
	7,960	7,300	9,860				

* Count C in this group of experiments was complicated by the injection of colchicine 16–17 hr. before the count was made. See text.

C = Cortrophin, dosage in Sudanophobic units (s.u.).   E = Eschatin, dosage in dog units (d.u.).

Table 2. *Blood lymphocytes per cu.mm. (ordinary type), red blood cells in 1000's per cu.mm. (black type) and reticulocytes after daily subcutaneous injection into rats of Cortrophin* (C), *Eschatin* (E) *or Cortrophin + Eschatin* (C + E)

Count A immediately before injections commenced.
Count B 27–28 days after first injection.

| No. of animal	Count A	Count B	Reticulocytes % Count A	Reticulocytes % Count B	Time between count A and B (days)	Substance used and daily dose per 100 g. body weight*	Weight in g. Initial	Weight in g. Terminal
44	6,703	9,854	0·0	3·2	28	C—3·0 s.u.	333	292
	7,460	7,500						
45	6,612	8,595	0·4	6·4	27	C—1·5 s.u.	329	292
	8,120	6,840						
46	6,974	10,999	1·2	4·8	28	C—4·5 s.u.	324	350
	8,840	7,720						
48	15,742	9,397	1·6	3·8	28	C+E—1·5 s.u. 1·0 d.u.	239	246
	10,060	8,800						
51	16,700	12,822	1·8	3·1	27	C+E—3·0 s.u. 1·0 d.u.	238	257
	7,900	9,004						
54	12,205	9,507	2·2	5·2	27	E—0·5 d.u.	230	256
	7,600	8,120						
55	15,506	9,800	2·5	4·2	27	E—1·0 d.u.	245	250
	8,140	8,040						
56	13,117	9,330	3·5	5·9	27	E—2·0 d.u.	251	287
	8,320	7,960						

* For explanation of terms see Table 1.

daily from 16 April to 26 May 1940, the blood lymphocytes were: 1 May, 3860 per cu.mm.; 20 May, 3180 per cu.mm.; and 27 May, 3620 per cu.mm. Again these counts were of the same order as counts performed during a preceding control period. It will be seen, then, that continued administration of cortical extract in the two rabbits had no marked effect on the lymphocytes in the peripheral blood.

HISTOLOGICAL OBSERVATIONS

Colchicine. As far as concerns the action upon lymphoid tissue of PATH and cortical extract, the experiments in group I were invalidated by the use

by Simpson *et al.* (1943). But the lessened amount of lymphoid tissue which is present seems quite normal, both in the cortex and the medulla, and mitoses could be observed in the germ centres (Pl. 2, fig. 6), though they did not appear to be very numerous.

Cortical extract. An unexpected result was obtained with cortical extract. As may be seen in figs. 7 and 8 (Pl. 2), the lymphoid tissue of the nodes is not only not atrophied, but on the contrary appears to be more active than normal. The germ centres show numerous mitoses, and the cells are closely packed. The diffuse lymphoid tissue surrounding the germ centres is dense, and extends into the greater part of the medulla. The reaction,
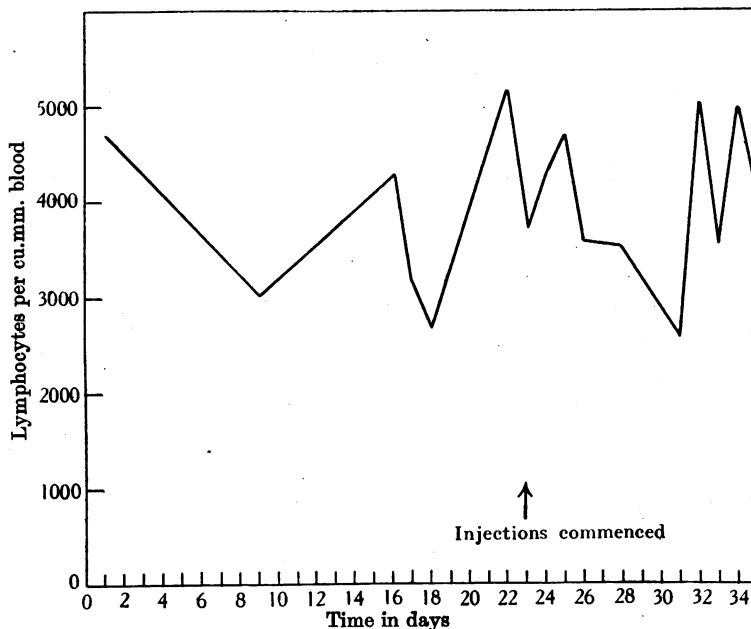
Text-fig. 1. Blood lymphocytes in rabbit following daily subcutaneous injections of cortical extract. Initial control period of 22 days. On the 23rd day, daily subcutaneous injections commenced of aqueous cortical extract, 12·5 dog units per kg. body weight.

of colchicine, which has a profoundly destructive effect on lymphoid tissue. Figs. 1 and 2 (Pl. 1) are low and high power views of a normal rat lymph node, figs. 3 and 4 corresponding views of a lymph node from an animal treated with colchicine. Fig. 4 shows the widespread pyknosis and karyorrhexis to which colchicine gives rise.

PATH. Figs. 5 and 6 (Pl. 2) illustrate the effect of the prolonged (4 weeks) administration of PATH on lymph nodes. The effect is quite definite if one compares these with the corresponding figures (Pl. 1, figs. 1 and 2) of the normal lymph node. The zone of cortical lymphoid tissue is greatly diminished in depth, while the medullary sinuses are dilated, giving rise to the appearance of 'flushing-out' noted

in fact, is a slight but definite hyperplasia. The presence of numerous macrophages is also evident even in a low power view (Pl. 2, fig. 7), both in the germinal centres and in the diffuse lymphoid tissue. They stand out as lightly-staining areas and give rise to the appearance of 'pitting' described by Dougherty & White (1945).

PATH + cortical extract. In two experiments in which PATH and cortical extract were given in combination (Pl. 3, figs. 9 and 10) the stimulating effect of the latter seems to have considerably neutralized the depressant effect of the former, as may be seen by comparing figs. 9 and 10 (PATH + cortical extract) with figs. 5 and 6 (PATH alone).

DISCUSSION

The differing reaction of lymph nodes to PATH and cortical extract

As has already been noted, the difference between the effect of PATH and of cortical extract was unexpected. Dougherty & White (1945) comment that degenerative changes in lymphoid tissue (mice and rabbits) 'were found within an hour following injection of either adrenotropic or adrenal cortical hormones and persisted for a 6 hr. period', and did not draw any distinction between the effects of hormone or cortical extract.

In the present observations, although the number of experiments was small (Table 2), the differences between the effects of the two substances was constant and definite (Pl. 2, figs. 5 and 7). These results are in conflict with those of other workers, but there are so many unknown factors involved that it is not possible satisfactorily to account for the discrepancy. There may, for example, be species differences. Thus Dougherty & White (1945) noted a marked difference between the reaction of lymphoid tissues in mice and rabbits. 'Within 24 hr., the lymphoid tissues of the mice resembled those in normal animals' (after giving PATH or cortical extract) 'except for the thymus which was still depleted of lymphocytes. In the rabbits, the degenerative changes recurred at 24 hr. after hormone injection.'

The dose and the time factor may also be important. In the present experiments the rats were killed 24 hr. after the last injection of hormone or extract, whereas Dougherty & White (1945) noted the maximal degenerative changes in mice within 6 hr. following injection. This would not explain in the present experiments the difference in reaction to PATH and cortical extract, though it may account for the failure to observe the more pronounced degenerative changes which they describe. For assuming that the immediate effect of cortical extract in the dosage employed is injurious, the lymphoid tissue may have time, if there is a sufficient interval between the doses of extract, and the damage not too severe, not only to recover but even to over-compensate for the injury and undergo hypertrophy. This type of lymphoid tissue reaction has been noted following the slight destructive action of small doses of X-rays (Drinker & Yoffey, 1940).

Another possibility is that PATH stimulates the cortex to secrete a much larger amount of hormone—or hormones—than is contained in the dose of cortical extract given. This, in the present state of our knowledge, can only be surmise, for we are not able to effect a quantitative correlation between the dose of PATH and the amount of cortical hormones secreted in response. In addition, there may be qualitative differences between the artifici-ally prepared extract and the natural hormones. In two experiments in which both PATH and cortical hormone were given in conjunction, the cortical extract inhibited the customary depressant action of the PATH (Pl. 2, figs. 5 and 6; Pl. 3, figs. 9 and 10).

The weight changes in the experimental animals (Table 2) introduce a further factor for consideration. In two PATH experiments (nos. 44 and 45) the animals lost weight, whereas in the third (no. 46) there was a gain in weight. In the remaining five experiments there was also a gain in weight. In all the animals of this series (Table 2) the general condition seemed excellent; the animals were lively and active, and took their food well. Nevertheless the loss of weight in nos. 44 and 45 is quite marked. From the work of numerous observers (see review by Jackson (1925)), it is clear that the lymphoid tissues are very sensitive to inanition and malnutrition. In the present experiments it may be significant that the atrophic changes in lymph nodes were more pronounced in experiments nos. 44 and 45, in which loss of weight was marked, than in no. 46, where the animal gained weight. It is worth noting that this animal (no. 46) received the highest dose of PATH.

Macrophages in lymph nodes. Dougherty & White (1945) described the appearance of increased numbers of large macrophages in both thymus and lymph nodes after the injection of PATH or cortical extract. 'The macrophages were so numerous that under low power the cortex of the thymus possessed a pitted appearance.' In the present experiments this appearance of 'pitting' was found in the rat thymus after PATH (Pl. 3, fig. 12) and cortical extract (Pl. 3, fig. 13) but also quite appreciably in the normal thymus (Pl. 3, fig. 11). It is also well marked in lymph nodes after cortical extract (Pl. 2, fig. 7), and the macrophages then seem to stand out more clearly than in the normal node (Pl. 1, figs. 1 and 2; Pl. 2, figs. 7 and 8). Dougherty & White (1945) interpret the presence of these macrophages as being chiefly in response to the increased destruction of lymphocytes which they describe as occurring. But this interpretation is not altogether convincing. It is quite true that since Flemming (1885) first described these macrophages, and noted the stainable bodies which they contained (his 'tingible Körper'), it has been accepted by most workers that these are ingested lymphocytes. Pl. 1, fig. 2, and Pl. 2, fig. 8, illustrate clearly the presence of several macrophages and their stainable bodies. But in the present experiments the macrophage increase was most noticeable in the animals given cortical extract, even though there was no indication of increased lymphocyte destruction, with the exception of the animals which were given colchicine in addition. In these latter (Pl. 1, fig. 4) the usual karyoclasic phenomena were noted, whereas in all other animals (Pl. 1, fig. 2; Pl. 2, figs. 6 and 8;

Pl. 3, fig. 10) the overwhelming majority of the small lymphocytes appeared normal.

Furthermore, there are always some macrophages present in normal lymphoid tissue, and they become much more abundant when it is undergoing active proliferation. 'In practically all secondary nodules in the active stage there are a fair number of cells of macrophage character' (Bloom, 1938), and one might add not only in the secondary nodules, but also in the diffuse lymphoid tissue (see also Latta, 1921). In the light of these facts it is interesting to note that it was more especially in the rats which were given cortical extract alone that the macrophages were particularly in evidence, and it was precisely in these animals that the lymphoid tissue was unusually active, as shown by the large and packed germ centres, containing numerous mitoses, and the dense diffuse lymphoid tissue occupying most of the node (Pl. 2, figs. 7 and 8).

If the stainable bodies in the macrophages are ingested lymphocytes, as appears likely, it is probable that there are always a few degenerating lymphocytes even in normal lymphoid tissue, and their number increases with the increase in the total number of lymphocytes which occurs when lymphoid tissue undergoes active proliferation.

The level of the blood lymphocytes. The degree of activity of the lymphoid tissue might be expected to influence the lymphocyte content of the blood, and reference has already been made to the observation by numerous workers of lymphopenia after the administration of PATH or cortical extracts. The degenerative changes observed in lymphoid tissues seem to point to diminished lymphocyte production being the cause of this lymphopenia. In the terminology of Drinker & Yoffey (1940), where X is the number of newly formed lymphocytes entering the blood, and Y the number leaving the blood, the number actually present in the blood at any time depends on the balance between X and Y, and if $X/Y = 1.0$ then the level of the blood lymphocytes will remain constant. In the case of PATH and cortical extract, the lymphopenia would appear to be of the $X - /Y$ type, if there are frank degenerative changes in the lymphoid tissues. However, it is possible that this is not the only factor involved. Thus in the present experiments the lymph nodes after repeated injections of cortical extract seemed to be hyperactive, yet the blood lymphocytes showed a distinct drop (Table 2). In these circumstances X is more likely to be $+$ than $-$, but a lymphopenia would still be possible if Y increased to an even greater extent than X. Or briefly, the lymphopenia would be of the $X + /Y + +$ variety.

Dosage. Before concluding from any experiments with Path and cortical extract that the adrenal cortex regulates the activity of lymphoid tissue and the level of the blood lymphocytes, one has to consider how far the dose given corresponds with the secretion in the normal animal. As far as PATH is concerned, this question cannot yet be answered. But in the case of cortical extract the work of Vogt (1943) is of interest.

Vogt (1943) estimates the daily output of the cortex as equivalent to 2·3 c.c. of the particular cortical extract employed per 100 g. body weight, and states further that 'this figure is probably an underestimate'. The extract which she used cannot be directly compared with Eschatin, since methods of assay are different. But assuming that the extracts are of the same order of potency, the daily output of the suprarenal cortex would be equal to 115 dog units per 100 g. body weight per day. In the present experiments the dose of cortical extract given was very much smaller than this, ranging from 0·5 to 2·0 dog units per 100 g. body weight per day. This dose not only produced the changes in the lymphoid tissues and blood lymphocytes already noted, but also characteristic changes in the adrenal, such as the storage of birefringent material in coarse masses instead of the usual dust-like particles (Baxter, 1945). If Vogt's (1943) data are correct, it is surprising that these comparatively small doses should have such a definite effect.

Antihormones. In the experiments involving continued administration of PATH the question of antihormone formation should obviously be considered. We have at present no evidence to offer on this point.

SUMMARY

In healthy adult rats, and in the dose given, the subcutaneous injection of pituitary adrenotropic hormone (PATH) usually results in a lowering of the blood lymphocytes after 3–5 hr. Aqueous extract of suprarenal cortex has a similar effect. Continued daily injections of PATH or cortical extract or both in conjunction for a period of 4 weeks also usually tend to lower the blood lymphocytes, though the effect is more definite with cortical extract than with PATH. PATH given daily for 4 weeks produced regressive changes in lymph nodes and thymus, whereas cortical extract gave rise to a slight but definite hyperplasia of lymphoid tissue. Subcutaneous injection of colchicine is followed after 16–17 hr. by (a) widespread destructive changes in lymphoid tissue, (b) the appearance in the circulating blood of immature bone marrow cells, both myeloid and erythroid. In two rabbits, daily injections of aqueous solutions of suprarenal cortex for 16 and 40 days respectively produced no significant change in the blood lymphocytes.

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EXPLANATION OF PLATES

All sections fixed in Zenker-formol, embedded in paraffin, cut at 6μ, and stained iron haematoxylin. All taken from rats.

PLATE 1

Fig. 1. Normal lymph node. Cervical. × 50.

Fig. 2. Part of a germ centre and some surrounding lymphoid tissue from fig. 1. Note mitoses, and also large macrophages containing stainable bodies. × 600.

Fig. 3. Animal 33. Table 1. Cervical lymph node 16½ hr. after subcutaneous injection of colchicine (0·2 mg./100 g. body weight). × 50.

Fig. 4. Germinal centre and adjacent lymphoid tissue from fig. 3. Node seems disorganized, and most of the lymphocyte nuclei show pyknosis and karyorrhexis. The overall picture is one of typical karyoclasic shock. × 600.

PLATE 2

Fig. 5. Animal 45. Table 2. Mesenteric lymph node, from an animal given daily injections of PATH for 28 days. In comparison with the normal (fig. 1), note the diminution in lymphoid tissue, the narrow cortical zone with small and poorly developed germ centres, the medulla with large and dilated sinuses. × 50.

Fig. 6. Germinal centre, from fig. 5, with zone of small lymphocytes and a dilated lymph sinus adjoining. Note mitoses in germ centre. Small lymphocytes perfectly normal. × 600.

Fig. 7. Animal 56. Table 2. Cervical lymph node after twenty-eight daily injections of cortical extract. Note the dense lymphoid tissue and well-marked germ centres. Compare with fig. 1 (normal) and note the slight increase in 'pitting', due to larger and somewhat more numerous macrophages. Contrast also with fig. 5 (PATH) and note the marked difference in reaction of lymphoid tissue to PATH and cortical extract. × 50.

Fig. 8. Part of fig. 7. Note well-marked germinal centre, with numerous mitoses, and also several macrophages. This is a much more active germ centre than fig. 6 (PATH), and somewhat more active than fig. 2 (normal). × 600.
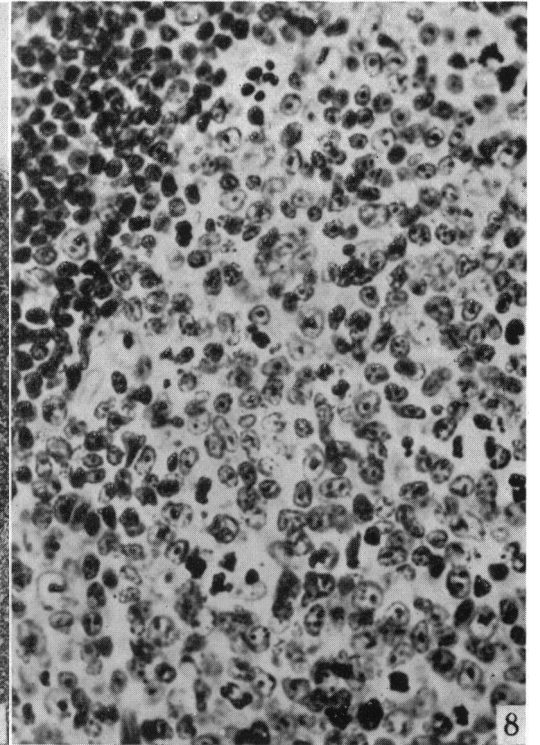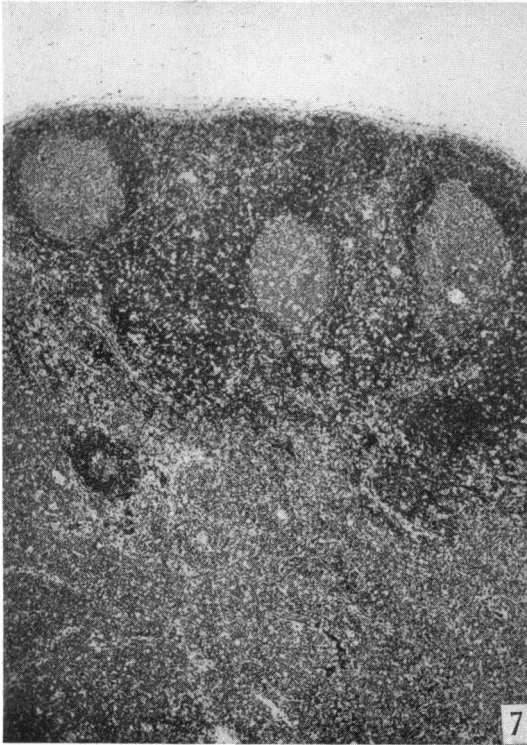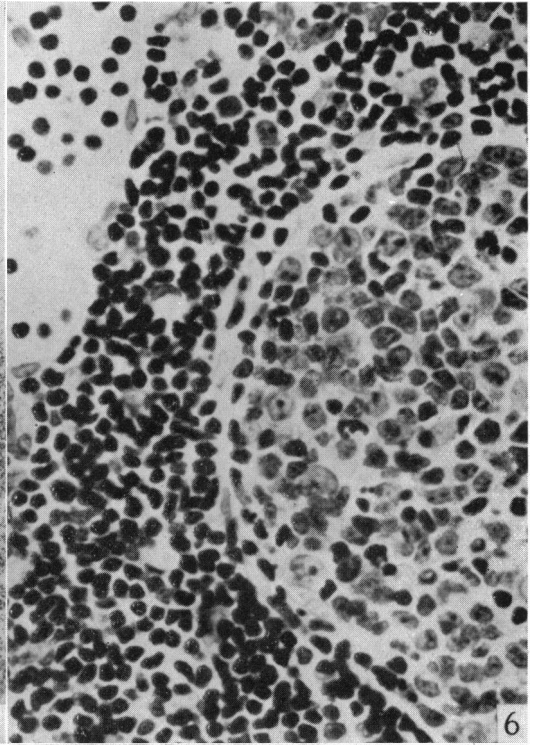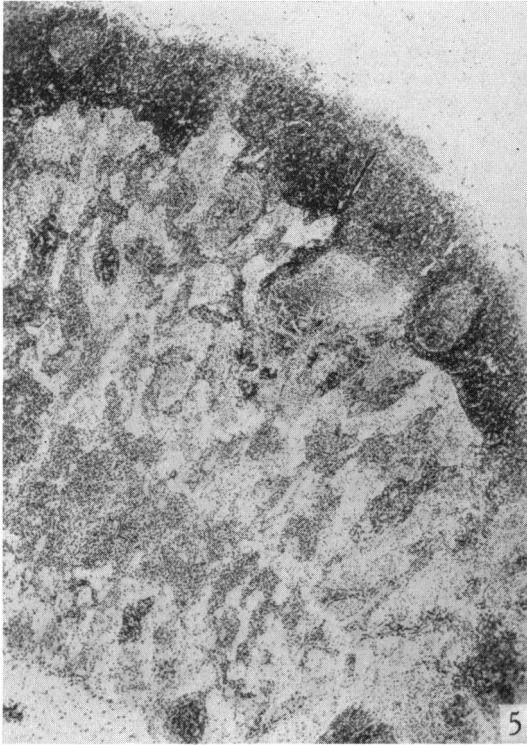
PLATE 3

Fig. 9. Animal 51. Cervical lymph node, from a rat injected with both PATH and cortical extract for 28 days. Compare with fig. 5, and note that the cortical extract seems to have diminished the effect of the PATH. × 50.

Fig. 10. Part of fig. 9. Note the active germ centre with numerous mitoses. × 600.

Fig. 11. Normal rat thymus. Note the 'pitting' normally present. × 50.

Fig. 12. Animal 46. Thymus of rat injected with PATH for 28 days. Compare with fig. 11 and note the diminution in cortical area, and increase in medulla. × 50.

Fig. 13. Animal 56. Thymus from rat treated with cortical extract for 4 weeks. Cortex dense, packed with small lymphocytes. Compare with fig. 12. × 50.
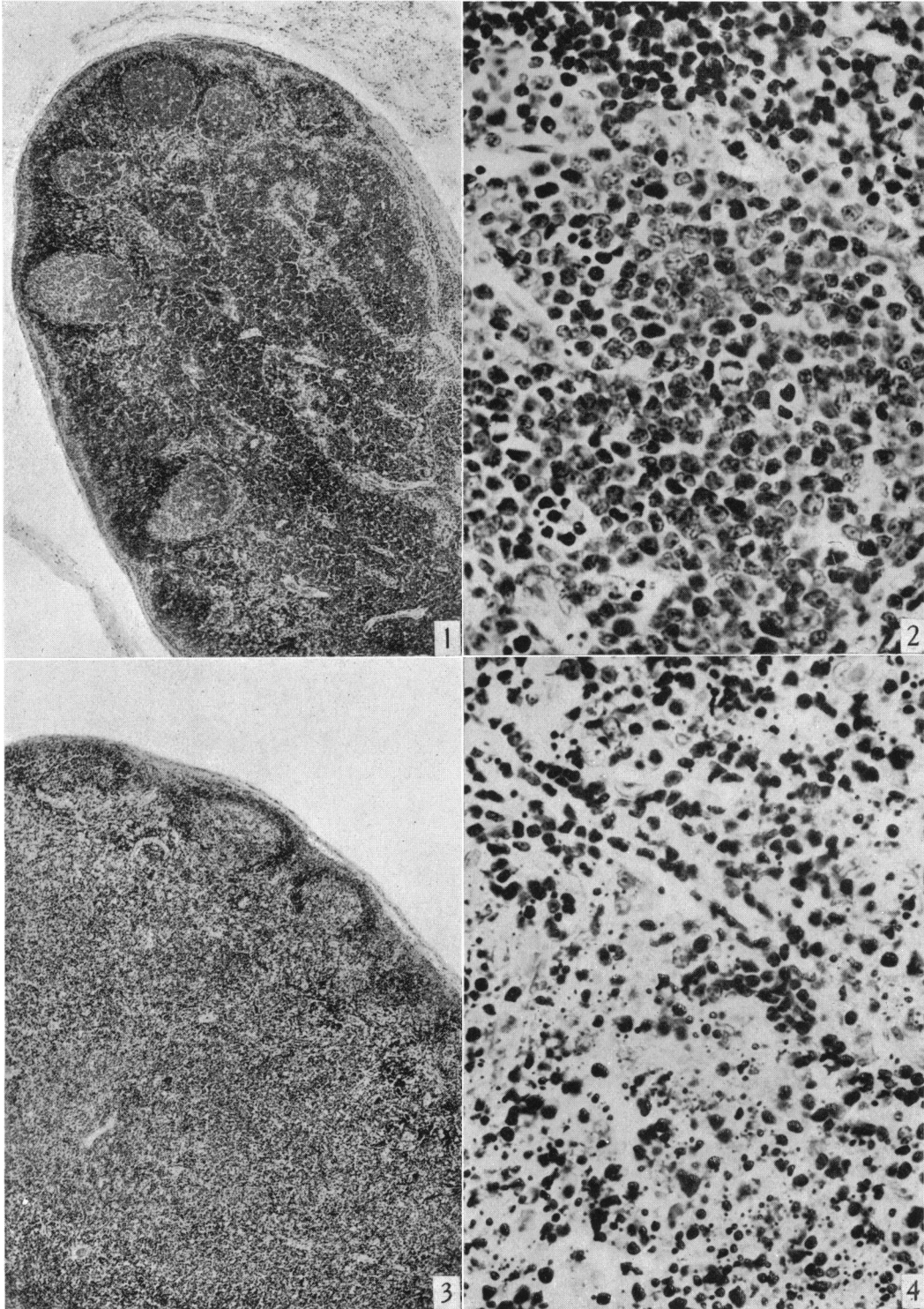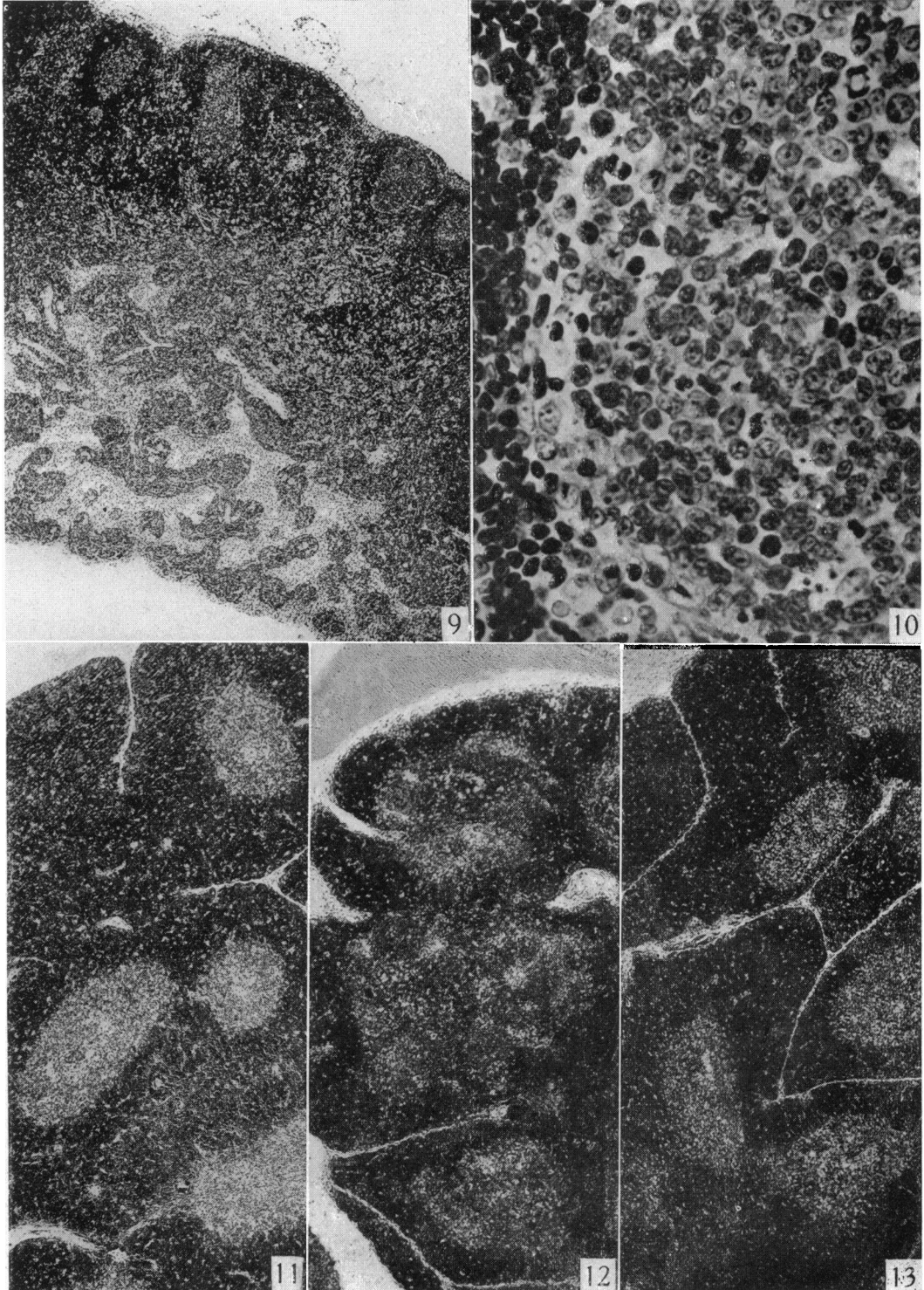
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