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SOME EFFECTS OF CORTICOSTEROIDS ON TISSUE HISTAMINE LEVELS IN THE GUINEA-PIG

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Available evidence from several species of animals points to an effect of corticosteroids whereby the biosynthesis, storage, catabolism and general tissue sensitivity to the effects of histamine may be modified or controlled. Of particular interest are the effects of these steroids upon tissue histamine levels since this may relate to the quantities available for release during conditions of inflammation or hypersensitivity.

In the rat, treatment with adrenocortical steroids causes a fall in histamine levels of many tissues (Hicks & West, 1958a; Telford & West, 1960; Cass & Marshall, 1962), whereas in adrenalectomized rats an increase in tissue histamine levels occurs (Rose & Browne, 1941; Marshall, 1943; Hicks & West, 1958b; Telford & West, 1961).

Similar effects of corticosteroids in the guinea-pig have been indicated by Kovacs (1965) who has shown that prolonged administration of cortisone results in a fall in levels of tissue histamine, whereas prolonged treatment with metyrapone, an inhibitor of adrenocortical secretion, causes an increase in tissue histamine levels. It was also indicated that simultaneous administration of corticotrophin and metyrapone may result in a further increase in tissue histamine. The latter result suggests that the action of metyrapone may not be explained simply on the basis of adrenal deficiency.

An investigation has been performed on the long- and short-term effects of both glucocorticoid and mineralocorticoid substances on tissue histamine levels in guinea-pigs, in which further aspects of these actions have been explored.

METHODS

Groups of five female albino guinea-pigs (Dunkin Hartley strain), weighing 300 to 500 g, were used in all experiments.

Following the administration of corticosteroids or related substances, tissue histamine was determined by the method of Parratt & West (1957). Samples of lung and ileum were taken from each animal in test and control groups, each sample being extracted with 10% trichloracetic acid and assayed for histamine on the atropinized guinea-pig ileum preparation.

Intramuscular injections of the following substances were given either singly or repeatedly as indicated in the text:

Cortisone acetate injection B.P. (25 mg/ml.), dexamethasone 21-phosphate injection (4 mg/ml.), aldosterone injection (0.5 mg/ml.), deoxycortone acetate solution in arachis oil (10 mg/ml.), cortodoxone (Reichstein's substance S) solution in arachis oil (10 mg/ml.), fludrocortisone acetate (4 mg/ml.) suspension in 0.1% solution of Tween 20, and repeated subcutaneous injections of metyrapone ditartrate (100 mg/ml.). Control

groups of guinea-pigs received corresponding volumes of suspending fluid, saline, or arachis oil. Doses of corticosteroid chosen were those found to be optimal in modifying the anaphylactic reaction of guinea-pigs, or those approaching maximal tolerated levels.

RESULTS

Effects of single injections of corticosteroids on tissue histamine levels

Groups of guinea-pigs previously treated with a single intramuscular dose of cortisone (10 mg/kg body weight), dexamethasone (4 mg/kg) or deoxycortone (10 mg/kg), were killed at 6, 12, 18 and 24 hr after injection. Tissue histamine levels of lung and ileum were determined and compared with those from corresponding control groups. Similarly, the effects of single intramuscular injections of aldosterone (2 mg/kg) on lung and ileal tissue histamine were investigated 4, 8 and 12 hr after administration. Results are shown in Table 1.

TABLE 1

LEVELS OF HISTAMINE IN GUINEA-PIG LUNG AND ILEAL TISSUES AFTER TREATMENT *IN VIVO* WITH SINGLE INJECTIONS OF CORTICOSTEROIDS

Doses: cortisone, 10 mg/kg; dexamethasone, 4 mg/kg; fludrocortisone, 4 mg/kg; aldosterone, 2 mg/kg; deoxycortone, 10 mg/kg; and cortodoxone, 10 mg/kg. Results are means and standard errors for groups of five tissue samples. * $P<0.05$; † $P<0.01$

		Histamine level (μg/g) at time (hr)					
Treatment	Tissue	4	6	8	12	18	24
Cortisone	Lung	—	8·0±1·3	—	5·4±0·7	3·7±1·1	6·4±1·0
Control		—	6·7±1·1	—	5·9±1·2	4·2±0·5	6·3±0·8
Dexamethasone	Lung	—	5·9±1·7	—	9·3±0·7	4·4±1·1	4·8±0·6
Control		—	5·8±1·4	—	7·2±1·0	4·1±1·3	5·1±0·6
Fludrocortisone	Lung	6·4±1·1	—	9·3±1·0	5·1±0·7	—	—
Control		5·9±0·4	—	6·6±0·9	6·1±1·2	—	—
Aldosterone	Lung	†15·2±1·5	—	†13·4±1·3	8·2±1·4	—	—
Control		6·7±0·7	—	5·5±0·8	6·1±0·4	—	—
Deoxycortone	Lung	—	*9·8±1·6	—	8·4±1·7	5·3±1·6	5·8±1·0
Control		—	5·7±0·7	—	6·1±0·8	5·5±1·5	5·6±1·1
Cortexolone	Lung	*10·5±2·0	—	6·6±1·4	6·4±0·5	—	—
Control		5·4±0·8	—	4·1±1·0	7·2±1·2	—	—
Cortisone	Ileum	—	15·2±1·6	—	—	18·1±2·4	12·1±0·7
Control		—	12·0±1·8	—	—	14·8±0·8	14·4±1·3
Dexamethasone	Ileum	—	15·0±1·1	—	10·3±1·5	11·6±1·2	12·9±0·3
Control		—	14·6±2·0	—	9·9±1·8	13·3±1·0	15·6±1·4
Fludrocortisone	Ileum	17·9±3·3	—	12·5±1·0	11·0±0·5	—	—
Control		11·4±1·2	—	10·8±0·4	11·2±1·4	—	—
Aldosterone	Ileum	†24·9±2·7	—	12·3±1·1	11·8±1·6	—	—
Control		11·7±1·2	—	10·6±1·1	9·9±1·0	—	—
Deoxycortone	Ileum	—	18·9±1·8	—	10·9±1·4	18·2±2·4	16·6±0·8
Control		—	16·0±1·8	—	9·6±1·2	16·0±0·7	15·1±0·6
Cortodoxone	Ileum	15·7±2·6	—	†14·9±2·0	10·3±1·8	—	—
Control		10·2±1·2	—	9·9±0·6	9·6±1·0	—	—

It can be seen that single intramuscular injections of cortisone, dexamethasone or fludrocortisone produced no significant change in tissue histamine levels over a period of 24 hr. However, a significant increase in lung tissue histamine was observed 6 hr after administration of deoxycortone, and in animals treated with aldosterone and cortodoxone there was a significant rise in lung and ileal tissue histamine levels within 4 to 8 hr. The effects on ileal tissue histamine were smaller and of shorter duration.

Effects of prolonged repeated corticosteroid treatment on tissue histamine levels

Groups of guinea-pigs were treated repeatedly with daily intramuscular doses of either cortisone (10 mg/kg), dexamethasone (4 mg/kg) or deoxycortone (10 mg/kg). Groups receiving each form of treatment were killed 7, 14 and 28 days after commencement of treatment, and tissue histamine levels were estimated. Another group of animals which had received daily intramuscular injections of cortodoxone (10 mg/kg) for 28 days was also examined. Comparison of results with those from control animals is shown in Table 2.

TABLE 2

LEVELS OF HISTAMINE IN GUINEA-PIG LUNG AND ILEAL TISSUES AFTER TREATMENT *IN VIVO* BY REPEATED ADMINISTRATION OF CORTICOSTEROIDS

Doses: cortisone, 10 mg/kg daily; dexamethasone, 4 mg/kg daily; deoxycortone, 10 mg/kg daily; and cortodoxone, 10 mg/kg daily. Results are means and standard errors for groups of five tissue samples. * $P<0.05$; † $P<0.01$

Treatment	Tissue	Histamine level (μg/g) at time (days) 7	14	28
Cortisone	Lung	5·0±0·6	6·1±0·7	3·5±0·8†
Control		6·9±1·7	7·4±0·5	7·2±0·6
Dexamethasone	Lung	7·8±0·7	6·4±0·8	3·5±0·2†
Control		7·4±0·5	7·4±0·4	6·8±0·4
Deoxycortone	Lung	5·9±1·8	8·1±1·1	6·9±1·1
Control		6·6±0·3	7·9±1·0	7·7±0·3
Cortodoxone	Lung	—	—	20·8±2·5*
Control		—	—	11·6±2·5
Cortisone	Ileum	12·7±0·9*	11·5±1·0*	8·8±0·9†
Control		15·5±0·8	14·5±0·6	13·9±0·9
Dexamethasone	Ileum	12·7±0·4	9·7±0·2†	7·3±0·7†
Control		12·6±0·2	13·6±0·5	14·1±0·4
Deoxycortone	Ileum	12·9±0·9	—	14·5±0·5
Control		14·7±0·8	—	13·4±0·7
Cortodoxone	Ileum	—	—	24·7±2·2*
Control		—	—	16·1±2·9

In groups treated with cortisone and dexamethasone a slow fall in lung and ileal tissue histamine levels occurred. This reduction became statistically significant only after 28 days of treatment. No significant change in histamine levels was seen in deoxycortone-treated animals, but there was a significant increase in the group which had received cortodoxone over 28 days.

Effects of repeated administration of metyrapone on tissue histamine levels

Several subcutaneous doses of metyrapone (200 mg/kg) were administered at 12-hr intervals to two groups of guinea-pigs. One group received two doses and was killed 12 hr

TABLE 3

LEVELS OF HISTAMINE IN GUINEA-PIG LUNG AND ILEAL TISSUES AFTER TREATMENT *IN VIVO* WITH METYRAPONE

Dose: 200 mg/kg, 12 hourly. Results are means and standard errors for groups of five tissue samples. * $P<0.05$

Treatment	Tissue	Histamine level (μg/g) after number of doses 2	7
Metyrapone	Lung	7·6±1·1	13·3±2·4*
Control		5·9±0·4	7·0±0·7
Metyrapone	Ileum	20·4±2·5	18·1±1·9*
Control		15·1±1·2	12·5±0·8

after the last dose, whilst the other group received seven doses and was killed 4 hr after the last dose. Lung and ileal tissue was taken from each animal and from saline-treated control animals. Histamine levels were determined and are shown in Table 3.

Metyrapone administration produced a rise in tissue histamine levels which was significant after 4 days of treatment.

DISCUSSION

The effects of the single doses of cortisone and dexamethasone appeared to produce no change in tissue histamine levels over 24 hr, and no change was observed over 12 hr in guinea-pigs treated with a single dose of fludrocortisone. However, prolonged daily treatment with cortisone and dexamethasone produced a gradual fall in tissue histamine levels. These results agree with those reported by Kovacs (1965).

In the guinea-pig the biosynthesis (Schayer, 1952), storage (Parratt & West, 1957) and enzymatic destruction (Lindell & Westling, 1953; Schayer, 1959) of histamine appears basically similar to that shown for the rat. In addition, there are some other indications of similar relationships between secretion of the adrenal cortex and the effects of histamine. For example it has been reported that, as in the rat, adrenalectomy increased the sensitivity of guinea-pigs to administration of histamine (Banting & Gairns, 1926) and decreased histaminase activity of some organs (Kahlson, Lindell & Westling, 1953). It is therefore possible that the reduction in tissue histamine levels resulting from prolonged administration of some corticosteroids depends upon a similar mechanism in both species. In the guinea-pig this decrease is somewhat slower than the comparable effect in the rat (Hicks & West, 1958a; Telford & West, 1960). In the rat the tissue turnover rate of histamine is faster than that in the guinea-pig; the estimated half-life of bound histamine in the guinea-pig is about 50 days (Schayer, 1952). Glucocorticoid treatment decreases the rate of binding of new histamine in rat tissues (Schayer, Smiley & Davis, 1954, 1956) and also decreases the histidine decarboxylase activity in the rat (Telford & West, 1961). The fall in tissue histamine levels seen with repeated injections of cortisone and dexamethasone may perhaps be explained on the basis of prevention of replenishment of tissue histamine, and the more gradual fall in tissue histamine levels of the guinea-pig related to the slower turnover rate in this species. A similar mode of action has been proposed for the decrease in histamine levels of guinea-pig tissues following treatment with derivatives of benzyl-l-isoquinoline (Parrot & Laborde, 1956), which were shown to depress histidine decarboxylase activity *in vivo*. In their experiments the rate of decrease in tissue histamine appeared to be greater than that observed with corticosteroid treatment, and may well have involved a mechanism other than passive removal by the normal turnover processes.

Administration of a single dose of aldosterone or cortodoxone led to marked but transient increases in lung and ileal tissue histamine within 12 hr, and a significant rise in lung tissue histamine occurred 6 hr after deoxycortone. In view of the apparent stability of tissue stores of histamine in the guinea-pig the relatively rapid onset of these effects contrasts strongly with the slow fall produced by cortisone and dexamethasone. A possibly analogous contrast is seen in the effects of adrenalectomy in the rat which produces a rise in tissue histamine levels at a rate relatively much more rapid than the fall in levels produced by cortisone administration. A possible explanation of the effect of these steroids is a brief

increase in the rate of formation of histamine, and from the transient nature of the effect it could be deduced that little of this histamine is bound in the tissues.

Kovacs (1965) showed that prolonged administration of metyrapone resulted in an increase in tissue histamine levels. Results presented in this paper indicate that this effect is apparent after a relatively short period of treatment with metyrapone.

Chart, Sheppard, Allen, Bencze & Gaunt (1958) described an inhibitory effect of metyrapone on the secretion of adrenal corticosteroids in the guinea-pig. Further work with this compound (Chart & Sheppard, 1959) indicated that this potent effect inhibited the secretion of all 17-hydroxycorticoids which in turn led to increased secretion of corticotrophin. As 11-β-hydroxylation reactions remained inhibited the cortex responded to corticotrophin by production of the 11-desoxysteroids cortodoxone and deoxycortone. It is suggested that the rise in tissue histamine levels effected by metyrapone in the guinea-pig is the result of inhibition of adrenal corticoid output, a mechanism analogous to the effects of adrenalectomy of the rat. It has been shown that prolonged treatment with cortodoxone results in a more persistent increase in tissue histamine levels than that produced by a single dose, although prolonged administration of deoxycortone produced no significant effect. However it may well be that the replacement of the normal adrenal cortical secretion by the mineralocorticoid cortexolone and deoxycortone could contribute to the effect of metyrapone on tissue histamine levels.

This suggestion is supported by the work of Kovacs (1965), who indicated that simultaneous administration of corticotrophin and metyrapone produced a further increase in levels of tissue histamine. This observation might be explained on the basis of a further increase in the output of 11-desoxysteroids.

Telford & West (1960) drew attention to the parallel activities of glucocorticoid substances in reducing tissue histamine levels of the rat, and their anti-inflammatory and anti-allergic properties. It is of interest to note that in some instances mineralocorticoid substances have been shown to exert a pro-inflammatory action (Selye, 1953; Selye & Bois, 1957; Ventura & Selye, 1957; Desaulles, 1958; Kellett, 1958, 1961; Bajusz & Jasmin, 1961). The increase in tissue histamine levels might thus be related to pro-inflammatory actions. Halpern (1956) and Halpern & Briot (1956) published evidence indicating that in histamine-depleted rats the pro-inflammatory mineralocorticoid deoxycortone increased the rate of replenishment of tissue histamine available for release by pharmacological agents. They were not, however, able to show any significant rise in histamine levels in rat skin after treatment with deoxycortone.

As with previous work in rats, the effects investigated have been produced by large doses of the various corticosteroids. It is thus difficult to explain these effects in terms of physiological mechanisms. It is also difficult to reconcile the relatively short onset and duration of most anti-inflammatory effects of corticosteroids with the slow decrease in histamine levels produced by repeated corticosteroid dosage. It may, however, be stated that, although no direct causal relationship may be deduced, the increased tissue histamine levels were associated with apparently pro-inflammatory mineralocorticoids while decreased tissue histamine levels were associated with glucocorticoids which have anti-inflammatory properties.

SUMMARY

1. Histamine in lung and ileal tissue of female guinea-pigs was extracted and assayed biologically after treatment *in vivo* with corticosteroids and related compounds.

2. Administration of single doses of aldosterone, deoxycortone or cortodoxone produced a large but transient rise in the histamine content of lung and ileum, but there was no change after cortisone, dexamethasone or fludrocortisone.

3. Prolonged administration of cortisone or dexamethasone over 14 to 28 days produced a gradual fall in tissue histamine levels, whereas cortodoxone, but not deoxycortone, increased histamine content.

4. Metyrapone increased lung and ileal tissue histamine levels.

5. The fall in tissue histamine levels produced by cortisone and dexamethasone may be a passive result of removal of histamine from tissues by normal turnover processes with accompanying inhibition of replenishment.

6. The effects of metyrapone may be the result of inhibition of normal adrenal cortical secretion, and a mineralocorticoid secretion may contribute to this effect.

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