Beta-Glycyrrhetinic Acid on the Adrenal Ascorbic Acid of Unstressed and Stressed Immature Female Rats

THE structural[1] and pharmacological[2–6] similarity between glycyrrhetinic acid and adrenal corticoids has been reported. Glycyrrhetinic acid resembles the corticoids in mineralocorticoid[2,3], anti-inflammatory[4,5], anti-œstrogenic[5] and anti-leukæmic[6] activity. Since the glycoside, glycyrrhizin, has an inhibitory action on the pituitary-adrenal system[7–9], it was of interest to ascertain whether the aglycone, glycyrrhetinic acid, exerted similar action.

In this investigation, adrenal ascorbic acid as a parameter of pituitary-adrenal activity was determined in both stressed and unstressed animals which had received short-term or prolonged administration of glycyrrhetinic acid. Beta-glycyrrhetinic acid (supplied by S. B. Penick Co., New York) was prepared in a vehicle of 1 part ethanol and 9 parts sesame oil. Immature *CFN* rats (23–24 days old and weighing 35–40 gm.) received subcutaneous injections of either 2 mgm. of glycyrrhetinic acid or an equal volume of vehicle (0·2 c.c.). Animals received three daily doses of the vehicle or drug as short-term administration. Prolonged administration consisted of 7 days of treatment with vehicle or drug according to the dosage schedule in Table 1. The specific stressor agent of 0·5 mgm./kgm. histamine was administered intraperitoneally 1 hr. before death. Animals were killed by decapitation. Both adrenals were cleaned, weighed and homogenized in 3 per cent metaphosphoric acid. The adrenal ascorbic acid was determined according to Bessey's method[10] in unstressed and stressed animals of both experimental groups.

Short-term administration of β-glycyrrhetinic acid (total dose 6·0 mgm.) had no effect on the response of the adrenal gland to the resting adrenocorticotrophic hormone (ACTH) secretory-rate nor did it impair the production of endogenous ACTH in response to a stressor. There were no differences between the ascorbic acid contents of unstressed treated and control animals nor stressed treated and control animals (Table 1).

Prolonged administration of glycyrrhetinic acid (total dose, 16·0 mgm.) significantly altered the adrenal ascorbic acid of the resting gland (Table 1). There was a significant increase (120·47 μgm./100 mgm.) in the adrenal ascorbic acid content of unstressed treated animals (Table 1). This might have indicated depression of adrenal corticoid output either through decreased ACTH secretion or decreased responsiveness of the adrenal gland. There was no evidence for the latter since there was no impairment of the pituitary-adrenal stress response (Table 1). The degree of adrenal ascorbic acid depletion in response to stress was the same in treated (31·43 per cent) and control (37·02 per cent) animals. The differences between the two groups remained the same, however (118·99 μgm./100 mgm.). The pituitary of treated animals was capable of responding to a stressor agent with an increased ACTH secretion equivalent to the controls and the adrenal gland was likewise capable of a response to increased endogenous ACTH. Prolonged administration of glycyrrhetinic acid therefore inhibited the secretory rate of the unstimulated pituitary. This inhibition was adequately overcome by the specific stressor histamine.

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Cholinergic Activity of Atropine

ATROPINE in suitable doses has been shown to potentiate the action of acetylcholine, and to possess anticholinesterase properties[1,2]. In addition, hyoscyamine is known to have cholinergic properties on the frog rectus muscle[3]. We have confirmed that atropine, in small doses, potentiates acetylcholine both *in vivo*, when the toxicity of acetylcholine is measured in mice, and *in vitro*, on the guinea pig ileum.

Groups of 5 male mice weighing 18–22 gm. were injected intraperitoneally with atropine 15 min. before receiving an intraperitoneal injection of

Table 1. EFFECTS OF GLYCYRRHETINIC ACID ON ADRENAL ASCORBIC ACID IN UNSTRESSED AND STRESSED IMMATURE FEMALE RATS

	Group 1*			Group 2†			Difference between groups 1 and 2	*P*
	Adr. asc. μgm./100 gm.	Diff. from control	*P*	Adr. asc. μgm./100 gm.	Diff. from control	*P*		
Unstressed control	(7) 490·40 ± 40·11			(11) 478·04 ± 30·66			12·30	N.S.
Treated	(6) 473·52 ± 35·27	−16·88	N.S.	(9) 598·04 ± 27·85	+120·47	< 0·01	125·52	< 0·02 > 0·01
Stressed control‡	(7) 347·78 ± 26·55			(11) 301·03 ± 27·11			36·75	N.S.
Treated	(7) 312·30 ± 22·65	−37·28	N.S.	(11) 410·02 ± 13·65	+118·09	< 0·001	97·72	< 0·001
Depletion control§	142·62 (29·08%)			177·01 (37·02%)				
Treated	162·22 (34·25%)	+19·60	N.S.	188·02 (31·43%)	+11·48	N.S.		

*Group 1: 23–24 day old rats treated for 3 days. Daily administration of 2 mgm. of glycyrrhetinic acid or 0·2 c.c. of vehicle subcutaneously. Total dose 6·0 mgm. killed on 4th day at 25–26 days of age.
†Group 2: 23–24 day old rats treated for 7 days. Daily administration of 2 mgm. of glycyrrhetinic acid or 0·2 c.c. of vehicle subcutaneously for 6 days. Double dose on 7th day. Total dose 16·0 mgm. killed on 8th day at 30–31 days of age.
‡Stressor: 0·5 mgm. histamine base /100 gm. body-weight intraperitoneal 1 hr. before killing.
§Depletion: Unstressed minus stressed. Per cent calculated as $\frac{\text{unstressed minus stressed}}{\text{unstressed}} \times 100$.

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