progress in order (1) to establish an exact incidence rate and (2) to elucidate the pathogenesis and functional significance of this interesting lesion with particular emphasis on the possible role of immunological factors.

Zusammenfassung. Spontane Schilddrüsenentzündung wurde in einer Gruppe von BUF Ratten beobachtet. Das histopathologische Bild der Schilddrüse wies grosse Ähnlichkeit mit Hashimoto's Schilddrüsenentzündung sowie experimentell hervorgerufener allergischer Thyroiditis in Laboratoriumstieren auf.

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Calcium Dependent Corticosteroid Release from the Perfused Cat Adrenal Gland

Calcium plays a crucial role in the secretion of many hormones and neurohumoral substances[1,2]. The intimate mechanism by which calcium regulates the secretory process is, at present, still a matter of conjecture; however, in most systems the secretory products are stored in granular or vesicular structures, and the release of the secretory products from these membrane-bound structures is thought to occur by the process of exocytosis[1]. It has been postulated that the action of calcium might be to facilitate an interaction between the granular and the cell membranes[2-4].

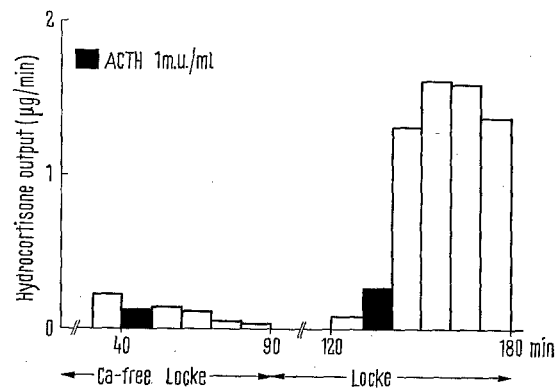
In the adrenal cortex, electron microscopic evidence indicates that secretory products do not appear to be stored in granular structures[5]. It is thought that during stimulation of the cortex by pituitary corticotrophin (ACTH), the steroids are synthetized de novo and then diffuse out to the cell exterior[6,7]. Therefore, an in vivo study to determine the importance of calcium on this secretory system might give further insight into the means whereby this divalent cation influences the secretory process.

Cat adrenal glands were perfused in situ through an aortic cannula at room temperature with normal or modified Locke's solution according to the method previously described[8]. The perfusate, collected from a cannula in the adrenolumbar vein, was assayed for 11-hydroxycorticosteroids according to the fluorometric method of MATTINGLY[9]. Since the major corticosteroid produced by the cat is hydrocortisone[10], outputs were calculated as µg hydrocortisone (alcohol) per minute.

When an adrenal gland was perfused for 40 min with Locke's solution from which calcium was omitted, no significant response to ACTH was observed (Figure). When the same concentration of ACTH was added some 60 min later but now in the presence of calcium, the response to ACTH was readily observable (Figure). The total steroid output during this 10 min exposure to ACTH plus the next 40 min in the calcium-free medium in the absence of ACTH was less than 10% of that observed in the presence of calcium (Table). When the calcium concentration was reduced to 0.2 m*M* (10% of normal), the response to ACTH was approximately one-half that of the control (2.0 m*M* calcium; Table).

Substitution of an equimolar concentration of strontium for calcium in the perfusion medium maintained the secretory response to ACTH (Table). However, magnesium (5 m*M*) was not able to replace calcium (Table). In fact, magnesium (15–20 m*M*) inhibited the ACTH-evoked steroid output in the presence of the normal calcium concentration (Table).

The present study indicates that the adrenal cortex and medulla manifest certain similarities in regard to their ionic requirements for secretion[8,11,12]. Thus, acetylcholine-evoked catecholamine output from the medulla is also drastically reduced in the absence of calcium; and strontium, but not magnesium can replace calcium. Furthermore, magnesium also antagonizes the effects of calcium in the medulla. These parallels are very interesting in light of the fact that the medulla contains a large quantity of preformed hormone which is sequestered within the membrane-bound chromaffin granules and is immediately released upon the initiation of the stimulus[8]. On the other hand, the action of ACTH on the cortex is thought to involve mainly an increase in steroidogenesis,



The release of hydrocortisone from the perfused cat adrenal gland by ACTH and its dependence on calcium. A gland was perfused initially with calcium-free Locke's solution for 90 min and ACTH (1 m.u./ml) was added during the 40th–50th minute of perfusion. Perfusion was then switched to Locke's solution containing calcium for an additional 90 min and ACTH (1 m.u./ml) was again added during the 130th–140th minute of perfusion. Each vertical column indicates the rate of steroid release in a 10 min period both in the presence and absence of ACTH.

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presumably through the action of cyclic 3′,5′-AMP[13], and this newly formed hormone diffuses out without being incorporated into the granular structure for storage[5].

Despite the presumed differences in the mode of secretion from the adrenal cortex and medulla, the requirement for calcium is readily evident in both structures. Thus, the question arises as to whether calcium has the same action in both endocrine organs. In vitro studies have shown that calcium is an important factor in the intra-mitochondrial synthesis of corticosteroids in response to ACTH[14,15]. The importance of calcium observed in the present studies and its relationship to adrenal mitochondria remains to be elucidated, and further studies are presently in progress to investigate this problem. However, it should be noted that in contrast to the present findings in vivo, strontium was a very poor substitute for calcium in the in vitro system[15–17].

The effects of divalent cations on the release of hydrocortisone from the perfused cat adrenal gland by ACTH

ACTH m.u./ml	Experiment	Steroid output[a] µg/50 min		Experimental as % of control
		Experimental	Control	
0.1	Ca^{+2}-free	0.11	3.59	3.1
1.0	Ca^{+2}-free	4.18	60.70	6.9
0.1	Ca^{+2} (0.2 m*M*)	17.65	37.60	46.9
1.0	Sr^{+2} (2.0 m*M*) Ca^{+2}-free	37.63	38.51	97.7
1.0	Sr^{+2} (2.0 m*M*) Ca^{+2}-free	18.53	15.44	120.0
0.1	Mg^{+2} (5.0 m*M*) Ca^{+2}-free	0.80	13.75	5.8
1.0	Mg^{+2} (15 m*M*) Locke	21.47	28.84	74.4
1.0	Mg^{+2} (20 m*M*) Locke	2.88	9.34	30.8

[a] Represents the total steroid output during a 10 min exposure to ACTH plus the next 40 min in the absence of ACTH. Control outputs were obtained in normal Locke's solution with the same concentration of ACTH as the experimental values which were obtained in the modified Locke's solution. Normal Locke's solution contained 2.0 m*M* calcium and 0.5 m*M* magnesium.

Zusammenfassung. Es wird gezeigt, dass die Freisetzung von ACTH in der durchströmten Katzennebenniere nur bei Anwesenheit von Kalzium erfolgt. Strontium, nicht aber Magnesium kann Kalzium freisetzen, während Magnesium (15 m*M*) die Freisetzung von ACTH in Gegenwart von Kalzium hemmt. Ein Vergleich zwischen Brenzkatechinamin und Kortikosteronfreisetzung wird diskutiert.

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Distribution of the Sulphur-35-Labelled Goitrogen L-5-Vinyl-2-Thio-Oxazolidone in the Rat

In earlier papers[1,2] we have demonstrated the high biological activity of the naturally occurring goitrogen L-5-vinyl-2-thio-oxazolidone (VTO). This substance we have been able to isolate from the seeds of cruciferous weeds growing abundantly in the endemic goitre districts of Finland and from the milk of the same districts[2,3]. In these papers the necessity of studying the distribution of this substance and its possible accumulation in the body, especially in the thyroid during long-term feeding, has been pointed out. This preliminary communication is concerned only with the distribution and delay of this compound in the body after one single dose.

Material and methods. The radioactive sulphur-35-labelled L-5-vinyl-2-thio-oxazolidone (L-VTO-S35) was prepared by the chemists of the Radiochemical Centre, Amersham, England, starting from the DL-aminobutenol (L-amino-3-buten-2-ol and 2-amino-3-buten-1-ol), separation of the L-isomer by resolution of its D-α-bromocamphor-π-sulphonate and heating of this substance with potassium hydroxide and carbon disulphide-S35, exactly as described by ETTLINGER[4]. The synthesis of the intermediate, DL-aminobutenol, was performed by Mr. A. ARSTILA, who primarily prepared 3,4-epoxy-1-butene (butadiene-1,2-oxide), which on aminolysis furnished the amino-butenols[5]. The purity of the substance was checked by IR-spectrophotometry.

L-VTO-S35 was given to rats as i.p. injection in 0.5 and 1.0 ml, respectively, of distilled water. The dose used was about 50 and 100 µg of VTO with a radioactivity of 11 and 22 µCi, respectively. The animals were killed under ether anaesthesia and the whole thyroid and suitable parts of liver and kidneys were weighed 6 h, and 1, 2, 4, 6 and 9 days after administration of the VTO dose. The radioactivity of the specimens was measured in a liquid scintillation counting equipment (Packard Tricarb) after homogenization of the tissue in

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