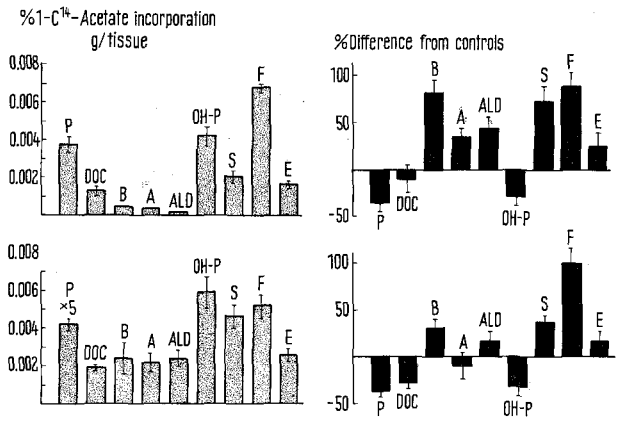
way(s) excluding free cholesterol and progesterone as intermediates. Therefore the site of action of angiotensin seems to be different from that of ACTH, which is supposed to act between cholesterol and pregnenolone[8,11,12].



Incorporation of 1-C^{14}-Na-acetate into corticosteroids and effects of angiotensin II (± S.E.M.).

Zusammenfassung. In Rindenschnitten frischer Rindernebennieren stimulierte Angiotensin II den Einbau von 1-C^{14}-Acetat in Mineralo- und Glukocorticosteroide, hatte jedoch keinen Effekt auf die Transformation von 4-C^{14}-Cholesterin und 4-C^{14}-Progesteron. Der Angriffspunkt des Angiotensins scheint danach in Synthesebereichen vor der Cholesterinbildung zu liegen.

D. LOMMER[13] and H. P. WOLFF

II. Medizinische Klinik und Poliklinik der Universität des Saarlandes, 665 Homburg (Germany), May 16, 1966.

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[13] Present address: Syntex Research, Institute of Hormone Biology, Stanford Industrial Park, Palo Alto (Calif., USA).

The Effect of Ouabain on Water Consumption in the Rat

In a recent publication from this laboratory, the paradoxical 'antidiuretic' effect of chlorothiazide in animals suffering from diabetes insipidus[1,2] was interpreted as an action on the thirst centre[3]. This explanation was derived from experiments with nephrectomized rats, in which water consumption was stimulated by hypertonic salt solution, the response being suppressed by chlorothiazide.

As this group of drugs interferes with active sodium transport in the kidney[4], it appears probable that they suppress thirst by a similar mechanism, viz. by curbing active sodium transport in hypothalamic (and other) osmoreceptors[3]. If this assumption is correct, any other drug that interferes with the functioning of the sodium pump should be capable of reducing drinking and thus should exhibit an antidiuretic effect. However, KENNEDY and CRAWFORD were unable to detect such an action with organic mercurials.

The inhibitory effect of digitalis glycosides on sodium transport is well documented[5,6] and serves as a basis for their diuretic action. The latter was unequivocally established by direct injection into the renal artery[7,8]. Therefore it was anticipated that these glycosides would affect the osmoreceptors regulating water intake. This prediction was verified with ouabain (*g*-strophantin), which is much more water-soluble than other cardiac glycosides and is not bound to serum proteins[9]. Therefore a rapid action can be expected.

Method. Rats of 200–300 g body weight were bilaterally nephrectomized under ether anaesthesia. Following the operation, they were kept for 24 h with food and water supply ad libitum. They were then injected subcutaneously with 50 ml/kg of 3% NaCl and their water consumption was measured. Groups of 4 rats received intravenous injections of a given dose of ouabain in isotonic saline, while an equal volume of saline was administered to the controls.

Results and discussion. Table I reveals a marked depression of water consumption in the ouabain-treated rats during the first 2 h. The effect then fades progressively. With the lower doses of the glycoside, complete recovery is indicated by the fact that the rats reach within 24 h the same level of water intake as the controls. However, above 0.5 mg/kg, ouabain shows a long-lasting effect which extends over more than 24 h (see Table I).

If % water consumption (control = 100%) is plotted against log dose, a straight line is obtained up to 1 mg/kg (Figure). For higher doses, the curve flattens out. In evaluating these results, it should be noted that spontaneous water intake is greatly reduced by nephrectomy. Thus in the present experiments normal rats consumed 130 ml/kg/h, while the operated animals drank only 51 ml/kg in the same period. However, injection of 3% NaCl raises the water intake of nephrectomized rats as much as in intact rats and sometimes even more. Thus in 5 series of experiments, normal animals drank 55 ml/kg of water during the first 2 h after administration of hypertonic saline, while the operated rats consumed 61 ml/kg. It should be recalled here that ouabain is rapidly excreted by the kidneys, but this route is blocked in the present procedure. Therefore we have also determined the toxicity of the glycoside in nephrectomized rats (see Table II). It is evident that in the operated animals

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ouabain is less toxic than in intact rats. This curious phenomenon is now under investigation. The LD_{100} of intact animals has been determined previously as about 17 mg/kg[10] and the LD_{50} as 5.5 mg/kg[11]. Variations in the toxicity of ouabain in different strains of rats is not unusual.

In the experiments with chlorothiazide[3], nephrectomy was necessary in order to eliminate interference by the diuretic action of the dosage applied. However, pronounced oligodipsia is found in nephrectomized rats with doses of ouabain only about $^1/_{25}$–$^1/_{50}$ of the LD_{50} (see Figure). Such doses are unlikely to exert any diuretic action. Therefore it appeared possible that the oligodipsic effect of the glycoside may be demonstrated also in intact animals. The Figure shows that indeed normal rats drink less when small doses of ouabain are administered. The dose-response curve is less steep than the curve for nephrectomized animals, and in the range of 0.25 to 2.0 mg/kg the effect on normal animals is weaker.

The present observations reveal a new extrarenal effect of ouabain, antagonistic to its diuretic action, but do not yet permit safe conclusions as to its localization. Further experiments with implantation of ouabain into various parts of the brain are necessary to establish beyond doubt a central mechanism for the oligodipsia evoked by this drug.

Table I. Effect of ouabain on water intake after subcutaneous injection of hypertonic sodium chloride

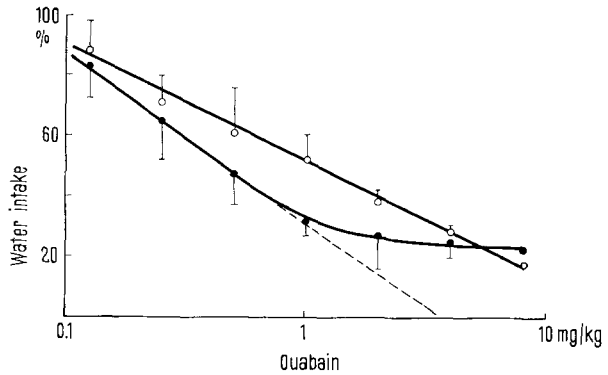
Ouabain mg/kg	Water consumption (ml/kg) during the period (h)				
	0–2	2–4	4–20	Total	% of control
A. Nephrectomized rats					
Control	49	25	53	127	100
0.25	36	31	54	121	95
0.5	26	21	50	97	76
1.0	16	8	40	64	50
B. Normal rats					
Control	58	11	75	144	100
0.25	45	9	93	147	102
0.5	46	9	74	129	90
1.0	28	7	65	100	69

Groups of 4 rats received 50 ml/kg of 3% NaCl by subcutaneous injection and ouabain, dissolved in 0.5 ml isotonic saline, intravenously. The controls received intravenous saline only

Table II. Acute toxicity of ouabain in normal and nephrectomized rats

Ouabain mg/kg	Death rate	
	Normal rats	Nephrectomized rats
9	0/4	
10	2/8	
11	2/8	0/4
12	7/9	1/8
13		2/4
14		6/7

Groups of 4 rats of 200-300 g body weight received i.v. injections of ouabain in 0.5 ml isotonic saline.



Dose-response curves for the depression of water intake by intravenous ouabain. Groups of 4 rats received 50 ml/kg of 3% NaCl s.c. and the dose of ouabain shown on the abscissa. o—o Normal rats; •—• nephrectomized animals. Each point, with the exception of 8 mg/kg, represents the mean of 3 experiments. Vertical bars indicate standard deviation.

Résumé. L'ouabain diminue la consommation d'eau, stimulée par l'injection d'une solution sallée hypertonique, chez les rats normaux ou néphrectomisés.

F. Bergmann, A. Costin,
M. Chaimovitz, and F. Benzakein

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Chronic Hepatic Injury Following Experimental Viral Hepatitis in the Dog

Experimental production of chronic hepatic disease in animals has been possible only by withdrawal of essential nutrients, administration of toxic chemicals or a combination of both. Although much useful information has been obtained by these methods, the relation of hepatic injury produced in this way to the chronic liver disease seen following viral infection remains doubtful. The availability of an experimental model bearing a closer resemblance to chronic hepatic disease in man would be desirable. This report describes a spontaneously occurring form of prolonged hepatic injury in dogs following experimental infection with the virus of hepatitis canis. This disease pattern, when contrasted with the course of acute canine hepatitis, appears to be distinctly different.

In the course of a study on the correlation of viral invasion with hepatic dysfunction[1] during hepatitis in 12–16-week-old pure-bred Beagle dogs, at least 16 examples of subacute and chronic progressive hepatic damage have been encountered. These puppies of 5–7 kg

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