rect antagonistic action of aldosterone, but as a result of the potassium depletion.

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NaCl Appetite of Adrenalectomized Rats.* (23670)

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One of the striking behavioral characteristics of adrenalectomized rats is their spontaneous NaCl appetite when given choice between water and .15 M NaCl solution as drinking fluids(1,2). This characteristic has been used by some investigators to assess completeness of adrenalectomy(3,4). It was therefore deemed worthwhile to determine whether the NaCl appetite of adrenalectomized rats was concentration dependent; that is, whether the rats preferred NaCl solution to water at some but not all concentrations of the former. It was also the object of this study to determine the NaCl concentration at which adrenalectomized rats ingested most salt solution. Knowledge of this particular concentration would be especially useful in experiments in which completeness of adrenalectomy is to be assessed, since it would assure that maximal differences could be obtained between intakes of normal and adrenalectomized rats.

Methods. Two separate experiments were performed. The materials and methods, similar for both experiments, are described below. Male rats of the Holtzman strain were used. All rats were kept in individual cages in a room maintained at 26 ± 1°C and illuminated from 8 a.m. to 6 p.m. The animals were allowed Purina Laboratory Chow *ad libitum.* Intakes of food were not measured. Each rat was given choice of 2 drinking fluids, distilled water and NaCl solution. The type of fluid containers used has been described (5). The positions of the 2 bottles were interchanged daily to avoid habit formation in the selection of the fluids. Fluid intakes were measured daily by weight difference and are expressed as ml/100 g body weight/day. Body weights of the rats were also measured daily. *Exp. 1.* Five adrenalectomized and 5 control rats ranging in initial weight from 180 to 200 g were used. Adrenalectomy was performed one week prior to beginning any measurements, and all animals were maintained on .15 M NaCl solution and water until the experiment began. Each of 4 different concentrations of NaCl solution was offered, with distilled water as the second fluid choice, for 4 days. Chronologically the concentrations given were: .25, .35, .50 and .15 M. *Exp. 2.* Five adrenalectomized and 5 control rats ranging in initial weight from 220 to 240 g

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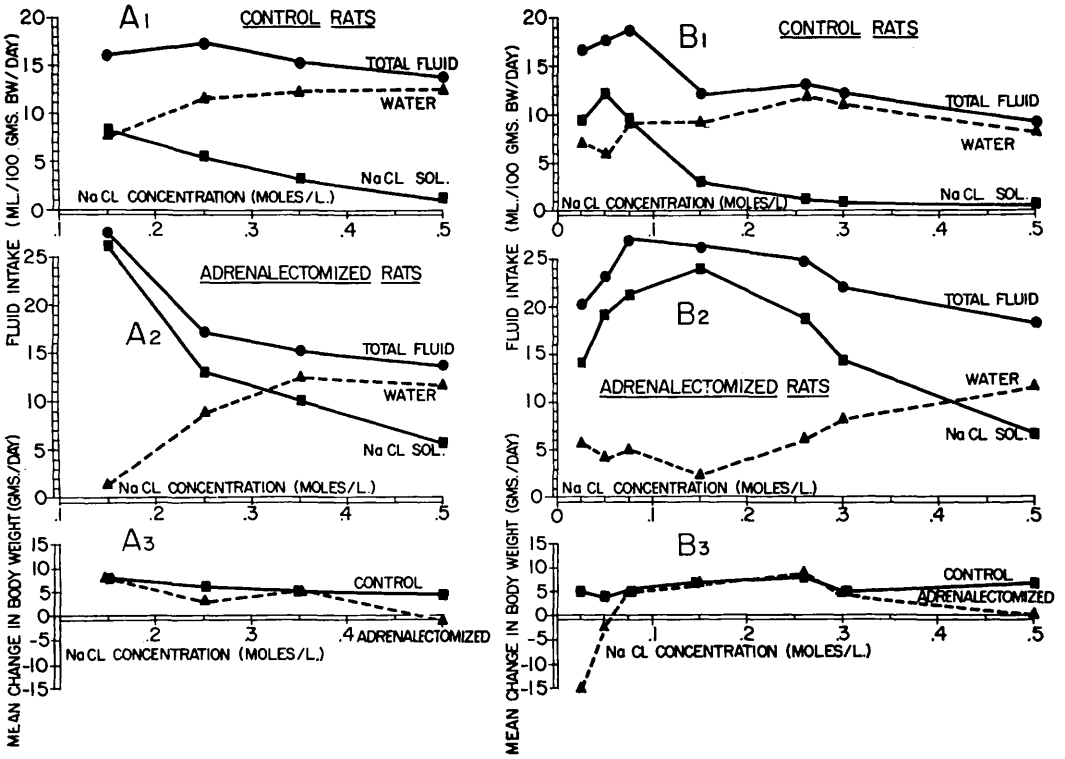

FIGS. 1A and 1B. Water, NaCl and total fluid intake of control (A_1 and B_1) and adrenalectomized (A_2 and B_2) rats in Exp. 1 and 2 respectively. Changes in body weight of both groups of rats in each experiment are shown in A_3 and B_3.

were used in this study. They were not the same animals used in the first experiment. Adrenalectomy was performed one week prior to the beginning of the experiment. All rats were given choice between water and each of 7 different concentrations of NaCl solution. This experiment differed from the first in that the rats were offered choice between a test concentration of NaCl and distilled water for one day and then were given choice between .15 M NaCl solution and water for 2 days Choice was then given between the second test concentration and water for one day followed by 2 more days with choice between .15 M NaCl solution and water. This pattern was followed throughout the experiment to prevent the rapid decreases in body weight of adrenalectomized rats accompanying low NaCl intake. Chronologically the test concentrations of NaCl given were: .15, .075, .050, .025, .26, .30 and .50 M. At the end of each experiment, completeness of adrenalectomy was assessed by removal of the salt solution only. Rats from whom adrenals have been completely removed do not survive longer than 15 days without supplementary NaCl(6). Completeness of adrenalectomy was also assessed by gross inspection for adrenal tissue at autopsy. Statistical analysis of the differences between means was performed by the use of the "t" test for the 5% confidence limit(7).

Results. The results of Exp. 1 are shown in Fig. 1A. It may be observed that control rats ingested similar ($P>.05$) volumes of salt solution and water when the test concentration was .15 M. When the NaCl concentration increased to .250 M, the volume of NaCl solution ingested decreased significantly ($P<.05$) below the simultaneous water intake (Fig. $1A_1$). Total fluid intake remained relatively constant but decreased when the highest concentration was given (.50 M). Adrenalectomized rats ingested over 3 times as much .15 M NaCl solution as control rats (Fig. $1A_2$). As the concentration of the NaCl

TABLE I. Mean Fluid Sodium Intake of Adrenalectomized and Normal Rats at Each NaCl Concentration Offered.

	NaCl concentration							
	.025 M	.050 M	.075 M	.15 M	.25 M	.30 M	.35 M	.50 M
Exp. 1	Mean fluid Na intake (meq/100 g body wt/day)							
Adx.				3.96	3.26		3.54	3.96
Control				1.25	1.38		1.10	.57
Exp. 2								
Adx.	.36	.96	1.63	2.71	4.86	4.31		3.36
Control	.24	.61	.72	.56	.29	.30		.38

solution increased, the volume ingested decreased markedly. When .35 M, or higher, NaCl solution was given, the adrenalectomized rats ingested significantly less salt solution than water. The total fluid volume ingested by adrenalectomized rats also decreased as the concentration increased. Control rats gained in body weight throughout the experiment while adrenalectomized rats showed a decrease in body weight only when .50 M NaCl solution was given (Fig. $1A_3$).

The results of Exp. 2 are qualitatively similar to those of Exp. 1 and are shown in Fig. 1B. With the wider range of NaCl concentrations offered, control rats drank most NaCl solution when the concentration was .050 M. At all concentrations above .075 M control rats ingested more water than salt solution (Fig. $1B_1$). Total fluid intake was highest when hypotonic concentrations were given (.025, .050, .075 M); relatively constant when .15, .26 and .30 M were given and lowest when .50 M was given. In the case of adrenalectomized rats, the largest volume of NaCl solution was ingested when .15 M NaCl solution was given. At concentrations higher than this, less and less NaCl solution was ingested. When .50 M NaCl solution was offered, significantly more water than salt solution was ingested. Total fluid intake of adrenalectomized rats also varied, decreasing as NaCl concentration increased.

Control rats gained weight throughout the experiment without regard to the concentration of the salt choice offered (Fig. $1B_3$). Adrenalectomized rats, on the other hand, lost weight when the 2 lowest concentrations were offered and failed to gain weight when the highest concentration was offered. The greatest weight gain occurred when .25 M NaCl solution was given.

Table I shows that the sodium taken in (meq/100 g B.W./day) by both adrenalectomized and control rats with their drinking fluids was relatively constant in the first experiment. The exception to this was the control group which decreased significantly ($P < .05$) its sodium intake below any of the previous levels when .50 M NaCl solution was given.

In the case of the second experiment in which a wider range of concentrations was offered, sodium intake was more variable in both groups of rats. The sodium intakes of adrenalectomized rats given .15, .25, .30 or .50 M NaCl solution did not differ from each other statistically ($P > .05$). However, sodium intakes of these rats given .025, .050 and .075 M NaCl solutions were significantly different from each of the higher concentrations. The variability in sodium intake from rat to rat was quite large. The sodium intakes of control rats were not different, one from the other, throughout the experiment. It is worth noting, however, that both groups of adrenalectomized rats ingested 2 to 17 times as much sodium by way of their drinking fluid as their respective controls.

When NaCl was removed from the cages of all rats at the end of both experiments, all adrenalectomized rats died within 10 days (mean 5.8; range 3 to 10 days). Autopsy of these rats did not yield adrenal tissue. It was therefore assumed that all the rats were completely adrenalectomized.

Discussion. The NaCl appetite of adrenalectomized rats has been shown to be concentration dependent in that less NaCl solution than water was ingested if the concentration of the NaCl solution was .35 M or greater.

The largest volume of NaCl solution was ingested when adrenalectomized rats were given .15 M. At this time the intake of NaCl solution represented 90 to 95% of the total fluid intake. A very large volume of .075 M NaCl solution was also ingested by adrenalectomized rats and consisted of 80% of the total fluid intake. On the other hand, control rats generally preferred water to NaCl solution except in low concentrations (.025 to .10 M). On the basis of this study, it would seem that .15 M NaCl solution is the best concentration to use for assessment of completeness of adrenalectomy. At this concentration the difference between intakes of salt solution of adrenalectomized and normal rats was maximal (Fig. 1).

The total fluid intakes of both groups of rats was greatest when hypotonic concentrations of NaCl solution were given (Fig. 1A and 1B). Conversely, the lowest total fluid intakes were observed when the highest concentration of NaCl was given (.50 M). It is difficult to explain from the data presented why both groups should take in more fluid in the hypotonic concentration range and less in the hypertonic concentration range.

In a multiple choice experiment, Young and Chaplin(8) gave normal and adrenalectomized rats simultaneous choice of 8 different NaCl solutions and reported that both groups of rats ingested most of their NaCl solution from the bottle containing a .12 M solution. The results of the single choice experiment reported here for adrenalectomized rats agrees with that reported by Young and Chaplin in that these rats ingested most salt solution when .15 M concentration was given. In the case of control rats, however, the results are different. The maximal volume of salt solution was ingested when .050 M was given. The different results may be due to the difference in experimental procedures; *i.e.* multiple *vs.* single choice.

Table I indicates that the pattern of the curves of both groups of rats in Fig. 1 cannot necessarily be attributed to attempts to maintain fluid sodium intake constant. When a wide range of NaCl concentrations was offered in Exp. 2, the sodium taken in by adrenalectomized rats did not remain constant. It is interesting that these rats should ingest nearly half as much water as NaCl solution when the concentrations lay below .075 M. At these concentrations one might expect the adrenalectomized rats to ingest salt solution to the exclusion of water, especially since adrenalectomized rats are known to have a preference ("taste") threshold several fold lower than that of normal rats which is known to lie between .008 and .016 M(9,10).

From the data presented, it cannot be stated accurately that the amount of sodium ingested with food remained constant. Under similar experimental conditions, however, it has been observed that the food intake, and therefore the food sodium intake, remained relatively constant (1.2 to 1.3 meq/100 g B.W./day)(10). It seems unlikely that the food intake of adrenalectomized rats could have varied to the extent necessary to maintain a constant sodium intake. Further study will be necessary to determine the factors influencing the pattern of the curves in Fig. 1.

Summary. In single-choice experiments adrenalectomized and normal rats were given choice between distilled water and each of a series of NaCl solutions. The NaCl appetite of adrenalectomized rats was shown to be concentration dependent. Adrenalectomized rats preferred NaCl solutions to water until the concentration of the salt solution was .35 M or greater. At these higher concentrations the rats preferred water to salt solution. Adrenalectomized rats ingested most NaCl solution (ml/100 g B.W./day) when the concentration was .15 M. These rats either lost weight or ceased to gain weight when low (.025 to .050 M) and high (.50 M) NaCl concentrations were given. Body weight of control rats was not influenced by the concentration of NaCl solution offered. Control rats preferred NaCl solution to water at all concentrations below .075 M. At concentrations greater than .15 M, the rats drank more water than NaCl solution. The greatest volume of NaCl solution (ml/100 g B. W./day) was ingested by control rats when a concentration of .05 M was given. The maximal difference between intakes of NaCl solution of control and adrenalectomized rats occurred when .15 M was given. Hence, this concentration

would appear to be best fitted to test completeness of adrenalectomy.

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Response of Individual Muscles of the Guinea Pig to "Large" Doses of Testosterone Propionate.* (23671)

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Administration of testosterone and several other androgens at various doses by multiple pellet implantation restored the individual muscles of the castrated male guinea pig only to normal(1). An attempt was made in this study to produce a response greater than normal by injection of "large" doses of testosterone propionate for a "long" period of time.

Procedure. Guinea pigs from the Zeimet Bio-farms were maintained in individual cages in an air conditioned room. They were fed Rockland Guinea Pig diet and supplements of 30 g of carrots and 25 mg of Vit. C twice per week. They were castrated at 68 days of age when they weighed 549 to 698 g. Injection of testosterone propionate† was begun on the day of castration and continued daily except Sunday for 65 days.

At autopsy the internal organs were weighed. The carcasses were sealed in plastic bags and kept frozen until the muscles were dissected.

Results. The castrated animals showed the expected(2,3) smaller gain in body (*cf* carcass) weight with the accompanying decreases characteristic of the respective muscles (Fig. 1). The 5 and 10 mg/day doses of testosterone propionate maintained all of the muscles except the temporal and digastric at or within the normal level. No significant differences between the 2 dose levels were detected. The 25 mg/day of testosterone propionate not only maintained many of the muscles at the normal level but also increased several above normal. These increases were less than 20% and occasionally as high as 30% (Fig. 1).

The seminal vesicles and prostates as well as the retractor penis did not show an excessive enlargement under the stimulation of the very high doses of testosterone propionate (Fig. 1).

The internal organs showed no remarkable changes in weight(1,2,3). The heart weight was slightly decreased after castration and maintained by the androgen. The thymus showed no change after castration but a slight decrease after androgen treatment. All of the other organs showed no changes.

Discussion. The guinea pig apparently possesses some mechanism(s) to protect it from excessive dosage of androgens. This phenomenon is not specific for any particular

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† The androgen was provided as perandren propionate by Ciba Pharmaceutical Products.