The area postrema does not modulate the long-term salt sensitivity of arterial pressure

J. P. Collister and J. W. Osborn

The area postrema does not modulate the long-term salt sensitivity of arterial pressure. Am. J. Physiol. 275 (Regulatory Integrative Comp. Physiol. 44): R1209–R1217, 1998.—The hindbrain circumventricular organ, the area postrema (AP), receives multiple signals linked to body fluid homeostasis. In addition to baroreceptor input, AP cells contain receptors for ANG II, vasopressin, and atrial natriuretic peptide. Hence, it has been proposed that the AP is critical in long-term adjustments in sympathetic outflow in response to changes in dietary NaCl. The present study was designed to test the hypothesis that long-term control of arterial pressure over a range of dietary NaCl requires an intact AP. Male Sprague-Dawley rats were randomly selected for lesion of the AP (APx) or sham lesion. Three months later, rats were instrumented with radiotelemetry transmitters for continuous monitoring of mean arterial pressure (MAP) and heart rate and were placed in individual metabolic cages. Rats were given 1 wk postoperative recovery. The dietary salt protocol consisted of a 7-day period of 1.0% NaCl (control), 14 days of 4.0% NaCl (high), 7 days of 1.0% NaCl, and finally 14 days of 0.1% NaCl (low). The results are reported as the average arterial pressure observed on the last day of the given dietary salt period: APx (n = 7) 114 ± 2 (1.0%), 110 ± 3 (4.0%), 110 ± 3 (1.0%), and 114 ± 4 (0.1%) mmHg; sham (n = 6) 115 ± 2 (1.0%), 114 ± 3 (4.0%), 111 ± 3 (1.0%), and 113 ± 2 (0.1%) mmHg. Neither group of rats demonstrated significant changes in MAP throughout the entire dietary salt protocol. Furthermore, no significant differences in MAP were detected between groups throughout the protocol. All lesions were histologically verified. These results suggest that the area postrema plays no role in long-term control of arterial pressure during chronic changes in dietary salt.

The role of the central nervous system in preserving normotension and homeostasis during chronic changes in salt intake remains to be fully understood. Aside from classic neural afferent pathways, much evidence now suggests that circumventricular organs (CVOs) can and do respond to changing levels of circulating hormones as an afferent signal(s) to modulate efferent neural outflow. This paper focuses on one such CVO, the area postrema (AP), as a possible relay site between circulating hormones and central sympathetic outflow.

The AP is a circumventricular organ of the fourth ventricle that lies on the dorsal surface of the medulla. Like other CVOs, the AP lacks the normal blood-brain barrier, and as such allows circulating substances, such as peptide hormones, access where they are otherwise excluded in the central nervous system (33, 34). In addition, the AP has been shown to have a rich supply of receptors for various hormones involved in cardiovascular regulation, such as ANG II, vasopressin, and atrial natriuretic peptide (17, 18). The AP is thereby thought to participate in neuroendocrine regulation by detecting circulating substances and transducing this information into neural messages (30).

Despite its other known functions, much research has focused on the function of the AP in autonomic control of cardiovascular function (1, 3, 17, 18, 21, 62, 66). For example, many studies have reported a role of the AP in mediating the sympathoexcitatory effects of ANG II. The idea of ANG II acting centrally was first proposed in the early 1960s (2). These and other studies were the first direct evidence of a sympathoexcitatory role of ANG II at the AP. These early experiments demonstrated that intravertebral arterial administration of ANG II produced a greater pressor response than did intracarotid or intravenous administration of ANG II in dogs and rabbits (22, 40, 50, 53, 55). Furthermore, this response was blocked in dogs after ablation of the AP (20, 35) and attenuated after ganglionic blockade (55). More recently, it has been shown that the chronic pressor effects of ANG II are mediated in part via the AP (13, 24) and that this chronic pressor effect is due to neurogenically driven vasomotor tone (13, 68). Further evidence of the role of the AP in modulating sympathetic activity is demonstrated by the fact that electrical stimulation of the AP causes changes in the activity of neurons in the rostral ventrolateral medulla, the site believed to be responsible for the generation of sympathetic activity (58, 63). Further review of the sympathoexcitatory role of ANG II at the AP is provided elsewhere (3, 19, 23, 35, 51, 57, 67).

In terms of the sympathetic nervous system itself and its relation to salt-dependent hypertension, many studies have implicated an impaired regulation of sympathetic activity linked to certain forms of salt-sensitive hypertension (5). A role of the sympathetic nervous system in the pathophysiology of salt-induced hypertension has been demonstrated in certain animal models, including the Dahl rat (25, 27, 39, 42, 54, 60), spontaneously hypertensive rat (SHR) (8, 36, 64), and deoxycorticosterone acetate-salt hypertensive rat (4) models of salt sensitivity. Additionally, Osborn et al. (49) demonstrated that when the sympathetic nervous system is effectively “damped” with the α1-adrenergic antagonist prazosin, a high level of salt-sensitive hypertension was produced in normal rats. Furthermore, in human patients, it has been shown that a high-NaCl diet decreases plasma norepinephrine levels in normo-
tensive and salt-resistant patients, but not in salt-sensitive patients (6, 16, 26, 37, 41, 45, 52). Alternatively, some studies suggest that increased salt intake stimulates sympathetic outflow in salt-sensitive subjects (38). It has been demonstrated that a high-salt diet causes a greater pressor response to norepinephrine in salt-sensitive patients compared with salt-resistant subjects (56). Although the relation between salt intake, the sympathetic nervous system, and salt sensitivity is not clear, all of these studies demonstrate a component of an inappropriately high level of sympathetic activity for the given circumstances. The underlying mechanism of how this relation is altered in salt-sensitive hypertension remains unclear.

If the sympathetic nervous system is in fact involved in the responses to changes in dietary salt, it also still remains unclear as to what is (are) the "signal(s)" to the central nervous system during such changes in salt intake. As previously mentioned, one possibility is that changing levels of circulating hormones could act as signals to the central nervous system via acting at CVOs. It is well known that many hormonal systems respond with altered levels of circulating hormones in response to changes in NaCl intake. For example, it is well established that the renin-angiotensin system (RAS) responds to changes in dietary salt content by altering plasma renin levels, and ultimately circulating ANG II concentrations. If in fact plasma ANG II can act at the AP to ultimately modify sympathetic activity, it seems plausible that this pathway may be involved in regulating sympathetic outflow in response to changes in dietary salt intake. In other words, when placed on a high-salt diet, suppression of the RAS would cause decreased levels of circulating ANG II. This in turn would be sensed as a signal at the AP to ultimately cause a decrease in sympathetic nervous system activity and maintain normotension. Therefore, in AP-lesioned (APx) rats, sympathetic nervous system activity would be inappropriately high because the signal for sympathoinhibition would be prevented by the lesion.

The present experiments were performed to test the hypothesis that an intact AP is necessary for the maintenance of normotension during chronic changes in dietary salt content. To test this hypothesis, we continuously monitored mean arterial pressure (MAP) and heart rate (HR) by radiotelemetry in intact and APx rats over a period of 8 wk. During this time, dietary NaCl intake was varied from low, normal, and high to examine the long-term salt sensitivity of arterial pressure.

**METHODS**

Adult male Sprague-Dawley rats (325–375 g, Harlan Sprague Dawley, Indianapolis, IN) were used in all experiments. All procedures were conducted in accordance with institutional and National Institutes of Health guidelines. Throughout the experiments, all rats were maintained in an environment with a 12:12-h light-dark cycle beginning at 7:00 AM.

**Surgical Procedures**

APx: Rats were randomly selected for APx or sham operation 12–13 wk before catheter implantation. Rats were preanesthetized with pentobarbital sodium (32.5 mg/kg ip) and atropine (0.2 mg/kg ip). Surgical anesthesia was achieved with a second intramuscular injection containing a combination of anesthetic agents (0.2 mg/kg acetylpromazine, 0.2 mg/kg butorphanol tartrate, 25 mg/kg ketamine). Rats were placed in a stereotaxic apparatus with the neck flexed. The surgical technique utilized for APx and sham operation was identical to that used previously in our laboratory (10, 11). Briefly, after midline incisions through the skin and epaxial musculature, the atlantocipital membrane was punctured and a portion of the base of the skull removed with rongeurs. At this point, with the AP clearly visible on the dorsal surface of the medulla, it was removed via suction by applying a blunt 26-gauge needle attached to a vacuum line. Sham surgeries were identical with the exception of the attached vacuum line. After APx or sham surgery, all rats were given an intramuscular antibiotic injection of 2.5 mg gentamicin and a subcutaneous injection of 0.075 mg butorphanol tartrate for analgesic purposes. All rats were then allowed 12–13 wk for postoperative recovery. The food intake of sham-operated rats was restricted to a level similar to that of APx rats during the first week of recovery. Food intake in the sham group was restricted to ~50, 60, and 80% of normal the first, second, and third weeks after sham surgery, respectively. After this initial period of food restriction, sham rats were allowed ad libitum access to food for the remainder of the recovery period. This length of recovery was chosen on the basis of our previous work examining the effects of food restriction and different periods of recovery in APx rats, demonstrating both normal MAP and HR in APx rats at this time point after surgery (11). In addition, we have previously reported that APx rats regain a normal food intake and growth rate 3 wk after the lesion (10). APx rats were allowed ad libitum access to food for the entire 12–13 wk of recovery.

Implantation of telemetry transmitter. After a period of 12–13 wk of recovery from APx or sham operation, rats were anesthetized as described above. Rats were then instrumented with radiotelemetric pressure transducers (model no. TA11PA-C40, Data Sciences International) for the purpose of continuous, 24-h sampling of MAP and HR. The unit consisted of a fluid-filled catheter attached to a transducer/transmitter. A midline abdominal incision was made to expose the descending aorta. After the aorta was clamped proximally, the catheter was introduced directly into the aorta via a 21-gauge needle. The catheter was advanced cranially so that the tip was just distal to the renal arteries. The catheter was glued in place with medical adhesive, and the aortic clamp was removed. The body of the transmitter was secured to the abdominal wall during closure of the body cavity with 3–0 silk sutures. The skin was closed with surgical staples. At the end of surgery, each rat received both an antibiotic and analgesic injection as described above. After recovery from anesthesia, rats were housed individually in metabolic cages (Nalgene). Rats were allowed 7 days to recover from surgery before the experimental protocol began. A 1.0% NaCl diet (Research Diets) and distilled water were provided ad libitum throughout this recovery period.

**Experimental Protocol**

The 8-wk experimental protocol was begun 7 days after telemetry/transducer implantation in two experimental groups: 1) APx rats (n = 7) and 2) sham rats (n = 6). The first 7 days of the protocol served as a control period during which time all rats were given a 1.0% NaCl diet (Research Diets). The dietary salt protocol continued as 14 days of 4.0% NaCl (high), 7 days of 1.0% NaCl (control), and finally 14 days of 0.1% NaCl (low) diet. At this time, to ensure all rats were in
sodium balance at this low level of sodium intake, all rats were given a daily injection of furosemide (1 mg/kg ip) for 2 days. The 0.1% NaCl (low) diet was then continued for another 14 days.

Throughout the protocol, MAP, HR, food intake, water intake, urine output, and body weight were measured daily in conscious, unrestrained rats in their home cages. MAP and HR were continuously monitored throughout the protocol using the Data Sciences telemetry data acquisition system (Data Sciences International). The arterial pressure signal was sampled at a frequency of 500 Hz for 5 s each minute. Each transmitter signal was monitored continuously by a receiver (model no. RA1010 or RLA1020, Data Sciences International) that was connected to a BCM 100 consolidation matrix (Data Sciences International). This matrix input was relayed to an IBM-compatible computer (ZEOS 486SLC) for analysis and storage. Data acquisition and analysis was performed using Dataquest IV software (Data Sciences International). Twenty-four-hour MAP and HR data were then calculated and stored for later analysis. Twenty-four-hour food and water intake as well as urine output were measured gravimetrically. Sodium intake was calculated as the product of urine flow rate and urinary sodium concentration. Confirmation of complete APx or intact AP (sham-operated rats) was made under light microscopy. All APx rats, there was minimal destruction of the adjacent nucleus of the solitary tract (NTS) at the microscopic level. Further evidence that APx did not impair NTS sites involved in the baroreceptor reflex was that 24-h lability of MAP (standard deviation of MAP), a quantitative index of baroreceptor reflex sensitivity (47), was not significantly different between APx (7.4 ± 0.2 mmHg) and sham (7.7 ± 0.2 mmHg) rats. Additionally, the 24-h lability of HR (standard deviation of HR) was not significantly different between APx (39.4 ± 1.5) and sham-operated (38.0 ± 1.7) rats.

Histological Verification of APx

Histological verification of APx was confirmed in all APx rats. A typical example is shown in Fig. 1. In all rats, there was minimal destruction of the adjacent nucleus of the solitary tract (NTS) at the microscopic level. Further evidence that APx did not impair NTS sites involved in the baroreceptor reflex was that 24-h lability of MAP (standard deviation of MAP), a quantitative index of baroreceptor reflex sensitivity (47), was not significantly different between APx (7.4 ± 0.2 mmHg) and sham (7.7 ± 0.2 mmHg) rats. Additionally, the 24-h lability of HR (standard deviation of HR) was not significantly different between APx (39.4 ± 1.5) and sham-operated (38.0 ± 1.7) rats.

Cardiovascular Responses to Chronic Changes in Dietary NaCl

MAP and HR were sampled every minute and then averaged for each 24-h period. Figure 2 illustrates an example of 24-h data from a sham and APx rat. The tracing begins at 7:00 AM (lights on) and continues for 24 h. The 24-h average and standard deviation for each tracing are shown. Note that the tracings appear similar in both magnitude and variability between the illustrated APx and sham rats.

Twenty-four-hour averages of MAP and HR throughout the protocol are shown in Fig. 3. Control MAP for the 7-day period of control (0.1% NaCl) diet was not different between groups (APx, 113 ± 2 mmHg; sham, 115 ± 2 mmHg). More importantly, no significant differences were seen in MAP between groups in response to the various changes in dietary salt throughout the entire protocol (Fig. 3A). The average MAP of each group observed on the last day of the given dietary salt period following the control period are as follows: 1) APx 110 ± 3 (4.0%), 110 ± 3 (1.0%), and 114 ± 4 (0.1%) mmHg; 2) sham 114 ± 3 (4.0%), 111 ± 3 (1.0%), and 113 ± 2 (0.1%) mmHg. Neither group demonstrated significant differences in MAP from control values throughout the entire protocol (Fig. 3A).

Although HR tended to be lower in APx rats throughout the 7-day control period, the average HR of each group during this period were not significantly different (APx, 345 ± 6 beats/min; sham, 356 ± 8 beats/min). We have previously reported significantly lower basal HR in APx rats that were recovered for the same period of time (11), although in that study parameters were measured only transiently for 10 min each morning. As shown in Fig. 3B, APx rats maintained significantly
lower HR than sham rats when dietary salt was increased to 4.0% and throughout the remainder of the protocol. The average HR of each group observed on the last day of each of the different dietary periods following the control period were as follows: 1) APx 332 ± 6 (4.0%), 332 ± 5 (1.0%), and 330 ± 6 (0.1%) beats/min; 2) sham 362 ± 6 (4.0%), 355 ± 5 (1.0%), and 352 ± 7 (0.1%) beats/min. Neither group demonstrated any significant change in HR from its respective control HR throughout the entire protocol (Fig. 3B).

Sodium and Water Balance Responses to Chronic Changes in Dietary NaCl

The 1-wk average of control sodium intake (1.0% NaCl) was not different between APx (2.8 ± 0.2 mmol/24 h) and sham (3.0 ± 0.1 mmol/24 h) rats. The average sodium intake of each group observed on the last day of each dietary NaCl period were as follows: 1) APx 10.6 ± 0.8 (4.0%), 3.2 ± 0.3 (1.0%), and 0.3 ± 0.0 (0.1%) mmol/24 h; 2) sham 12.8 ± 1.5 (4.0%), 3.7 ± 0.2 (1.0%), and 0.3 ± 0.0 (0.1%) mmol/24 h. Sham rats maintained significantly higher sodium intakes compared with APx rats during days 3, 5, 8, and 10–14 of the high (4.0%)-salt diet (Fig. 4A). Throughout the rest of the protocol (1.0% and 0.1% NaCl diets), no significant differences were seen between the groups (Fig. 4A).
In terms of sodium excretion, no significant difference in the average 7-day control values was seen between APx (2.1 ± 0.2 mmol/24 h) and sham (2.5 ± 0.2 mmol/24 h) rats. The average sodium excretions of each group on the last day of each of the given dietary periods were as follows: 1) APx 6.7 ± 0.9 (4.0%), 2.3 ± 0.2 (1.0%), and 0.3 ± 0.0 (0.1%) mmol/24 h; 2) sham 11.3 ± 1.0 (4.0%), 3.0 ± 0.2 (1.0%), and 0.4 ± 0.1 (0.1%) mmol/24 h. Significantly higher sodium excretions were seen in sham rats throughout the entire high (4.0%) salt diet (except day 6), and on day 1 of basal (1.0%) NaCl (Fig. 4B). No significant differences in sodium excretion were detected between APx and sham rats during the remainder of the protocol (Fig. 4B).

No significant difference was seen between the average water intake for the 7-day control period (1.0% NaCl) in APx (32.3 ± 2.7 ml/24 h) and sham (28.3 ± 2.0 ml/24 h) rats. The average water intakes of each group on the last day of each NaCl diet were as follows: 1) APx 44.2 ± 5.1 (4.0%), 36.7 ± 3.2 (1.0%), and 34.4 ± 3.9 (0.1%) ml/24 h; 2) sham 49.3 ± 4.4 (4.0%), 33.6 ± 3.3 (1.0%), and 29.3 ± 4.0 (0.1%) ml/24 h. No significant differences in water intake were seen between the groups throughout the entire protocol (Fig. 5A).

Although daily urine outputs during the control period tended to be greater in APx rats (Fig. 5B), there was no significant difference between the 7-day average control period between APx (16.3 ± 1.8 ml/24 h) and sham (11.8 ± 1.4 ml/24 h) rats. The average urine outputs of each group on the last day of each dietary period were as follows: 1) APx 26.1 ± 4.4 (4.0%), 19.6 ± 2.5 (1.0%), and 19.1 ± 3.4 (0.1%) ml/24 h; 2) sham 31.9 ± 2.1 (4.0%), 17.3 ± 2.0 (1.0%), and 15.3 ± 3.3 (0.1%) ml/24 h. No significant differences in urine output were seen between groups throughout the entire dietary salt protocol (Fig. 5B).
In addition to daily sodium and water balances, cumulative balances were also calculated for each group. No differences in cumulative sodium balance were seen between APx and sham rats during the 7-day control period (Fig. 6A). However, beginning on day 1 of high (4.0%) salt, APx rats tended to have a higher cumulative sodium balance compared with sham rats (Fig. 6A). This trend continued and was statistically significant by day 9 of the high (4.0%)-salt diet (Fig. 6A). The magnitude of this difference continued to grow through the end of this high (4.0%)-salt period. Significant differences in cumulative sodium balance between the two groups were maintained through the end of the protocol, but the overall degree of difference did not change after the high (4.0%)-salt diet and throughout the remaining two (1.0% and 0.1% NaCl) dietary periods (Fig. 6A). No differences in water balance were seen between APx and sham rats during the 7-day control period. Furthermore, no significant differences in cumulative water balance between APx and sham rats were seen throughout the entire protocol (Fig. 6B).

Responses to Furosemide

As reported in METHODS, each rat was given a daily injection of furosemide (1 mg/kg ip) at the end of the protocol for 2 days. This was done to ensure that all rats were indeed in a state of low body sodium content at the end of the protocol. All parameters previously measured were monitored for an additional 14 days while the rats were consuming the 0.1% NaCl diet. No changes in any parameters previously measured were detected (between or within groups) during this further 2 wk of low-salt diet (data not shown).

Fig. 6. Effect of chronic dietary salt changes on cumulative sodium (A) and water (B) balance in APx vs. sham rats. *Statistical significance between groups (P < 0.05).

DISCUSSION

The pathophysiology underlying salt-sensitive hypertension remains unclear. One approach to beginning to understand this problem is to address the following questions: 1) how is an increase in salt intake detected or “sensed” in the normal animal?; 2) what, if any, intermediate relay stations are necessary to convey this information?; and, last, 3) what effector mechanisms are then implemented to maintain arterial pressure and sodium homeostasis? With this approach, if these questions are addressed in the normal animal, certainly some of the underlying pathophysiological mechanisms of salt-sensitive hypertension can be further elucidated.

A neurogenic mechanism of salt-sensitive hypertension has been implicated in human hypertension (6, 37, 38) and many animal models of hypertension (5). More specifically, studies in SHR (8, 36, 64), Dahl salt-sensitive (25, 27, 39, 42, 54, 60), and prazosin-treated rats (49) all indicate a sympathetic component of the hypertension observed in these models. If the sympathetic nervous system is involved in responding to changes in salt intake, the question remains as to how this change is mediated or sensed by the central nervous system. Chronic changes in salt intake are known to cause changes in circulating hormones involved in renal control of sodium and water balance (e.g., ANG II, vasopressin, atrial natriuretic peptide). Interestingly, the AP has a rich supply of receptors for these same hormones and has been shown to mediate cardiovascular changes when exposed to certain of these hormones (17, 18). The obvious question is therefore could the AP be a primary central nervous system site for sensing changes in sodium intake via changes in levels of circulating peptide hormones, and in turn modulate sympathetic regulation of arterial pressure?

The present study was conducted to determine whether an intact AP is required to maintain arterial pressure during chronic changes in dietary salt consumption in the normal rat. In other words, is the AP an important mediating site in the prevention of salt-sensitive hypertension? In the present study, APx rats maintained normal arterial pressures throughout 14 days of high (4.0% NaCl), 7 days of normal (1.0% NaCl), and 14 days of low (0.1% NaCl)-salt diets. Furthermore, 24-h continuous arterial pressure measurements were made in these experiments using radiotelemetry transmitters. We have found this technique to be the most accurate during experiments utilizing chronic changes in dietary salt because often-times measurable changes in pressure only occur during the night, when the animals are consuming the salt (48). Throughout the protocol, these rats demonstrated no significant changes in MAP from control values and furthermore no significant differences in arterial pressure were seen between APx and sham-operated control rats. These results do not support the hypothesis that an intact AP is necessary to regulate arterial pressure during changes in chronic dietary salt consumption. Although the results of these experiments are in fact a “negative” finding, we feel these observa-
tions are important in the overall understanding of the AP and the long-term control of arterial pressure.

Much evidence has demonstrated an important role of the AP in the regulation of arterial pressure by sensing and responding to changing levels of circulating substances, such as peptide hormones (30). Furthermore, it has been postulated that the AP performs this task in part by modulating sympathetic outflow (3). Therefore, it seems plausible that the AP would play a role in the neurogenic control and maintenance of arterial pressure during changes in chronic dietary salt. For example, increased salt intake would cause decreased levels of circulating ANG II via suppression of the RAS. Subsequent lower levels of ANG II at the AP could then cause a neurally mediated suppression or withdrawal of sympathetic tone to maintain normal arterial pressure during periods of elevated salt intake.

The fact that APx animals did remain normotensive throughout wide changes in chronic salt intake leads to several interesting ideas. The first and most obvious explanation is that the AP is in fact not important in this aspect of cardiovascular and autonomic control. Consistent with this idea, we have recently reported that the AP is not necessary for the support of lumbar sympathetic nerve activity by endogenous ANG II in rats consuming a low-sodium diet (65). It is also possible that the AP does not play a role in cardiovascular regulatory responses to salt intake. The direct, nonneurogenic actions of changing concentrations of circulating hormones in response to dietary salt at other structures, such as the vasculature and kidneys, may be enough to prevent changes in MAP, in the absence of an AP. On the other hand, neurogenic actions of hormones at other central structures, such as the subfornical organ (18) or rostral ventrolateral medulla (59), may be the primary neural pathways involved in this homeostatic pathway. Furthermore, other salt-sensitive or osmoreceptor pathways involved in neural regulation may be the primary mediators in the prevention of salt-induced changes in MAP. Central osmoreceptors and more recently peripheral osmoreceptors (9) have both been postulated to be the afferent means of sensing changes in plasma sodium concentration as they relate to dietary salt consumption. These pathways have been shown to activate central hypothalamic nuclei (7), and as such could also be the primary means of regulating arterial pressure in the face of chronic changing dietary salt. Last, it is plausible to think that the AP does play a role in the maintenance of arterial pressure in the face of changing salt intake, but that other redundant pathways have compensated for the loss of the AP during the 3 mo of recovery after the lesion. This possibility is highlighted by the fact of numerous reports of conflicting cardiovascular results in the literature on APx rats, depending on the length of recovery before the given experiments were begun. For example, APx rats have been shown to have low basal MAP 3 wk postsurgery, but normal MAP at 3 mo after the lesion (10, 11). In fact, we have recently reported a different pattern of hypertensive responses to the AT\textsubscript{1} receptor antagonist losartan in APx rats recovered for 3 mo versus 3 wk (11). Therefore, it is possible that APx rats recovered for a shorter period of time might exhibit salt-sensitive hypertension when tested with this same protocol. This hypothesis remains to be tested.

In this study, basal HR values of APx rats were similar to sham rats during the 7-day control measurements. We have previously reported lower basal HR in APx rats recovered for the same amount of time (11), although in that experiment HR was measured transiently for 10 min during the day, in contrast to the 24-h continuous measurements made in this study. As such, this differential result could be explained by the difference in technique used between these two studies. By measuring 24-h HR in this study, elevated active, nighttime HR of rats were included in our analysis of the data. In this experiment, although the overall average HR in APx rats (345 ± 6 beats/min) was lower than shams (356 ± 8 beats/min), this difference was not statistically significant. Furthermore, APx rats demonstrated a bradycardic response during the high (4.0% NaCl)-salt diet that reached a nadir of ~335 beats/min by day 8 of this 4.0% NaCl diet. This low HR was maintained throughout the rest of the protocol and was unaffected by the further changes in dietary salt intake. On the basis of this last observation and our past experience, we suspect these animals began with slightly elevated HR and therefore this was not truly a bradycardic response to the experimental protocol. The fact that HR did not change (or increase) when the animals were again placed on the normal control (0.4% NaCl) diet supports this idea.

Last, in this study we saw no differences in cumulative water balance between sham and APx rats. This is reflected through equivalent daily water intakes and urine outputs seen between both groups. In contrast, APx rats demonstrated an increasingly higher cumulative sodium balance throughout the period of high (4.0% NaCl)-salt diet. Previous reports have demonstrated an increased salt appetite or consumption of saline solutions in APx rats (15, 32), which is apparently independent of the decreased food intake or anorexia observed in APx rats after surgery (61). The APx rats observed in this study demonstrated no increased sodium consumption and had similar daily food intakes as sham rats. Therefore, the higher cumulative sodium balance observed in APx rats in this experiment can be wholly explained by the fact that APx animals excreted less sodium, while overall actually ingesting slightly less sodium compared with sham rats during the high (4.0% NaCl)-salt diet. Clearly, in this study, APx rats had an impaired ability to excrete sodium when placed on a 4.0% NaCl diet. This suggests that APx rats have modified hormonal or neural control of renal function. It has been shown in renal-dener- vated rats (14, 46) and rabbits (28) that renal innervation is necessary to conserve sodium after dietary sodium restriction. These results do not correlate with our observations presented here, as APx rats in this study had no impairment in conservation of sodium. Interestingly, studies by Greenberg et al. (29) demonstrated an impaired ability of renal-denervated rats to excrete sodium when placed on high-sodium diets. These results demonstrated an increasingly higher
cumulative sodium balance in renal-denervated rats beginning ~24–48 h after being switched to a higher-salt diet (29). Likewise, similar to our observations at present, these studies showed no differences in arterial pressure between groups. Additionally, Nishida et al. (44) recently demonstrated that APx rabbits have an impaired ability to suppress renal sympathetic nerve activity and concomitant sodium excretion in response to a portal hypertonic saline load. Furthermore, we have recently reported that APx animals do in fact have higher basal lumbar sympathetic nerve activity compared with sham animals (65). These previous findings, in light of and similar to our current observations, could suggest that APx animals have a neurally mediated renal impairment of sodium excretion or an impaired ability to suppress renal sympathetic nerve activity. This suggests a role of the AP in the overall control of sodium handling by the body and neurally mediated sympathetic withdrawal to the kidneys during periods of sodium excess.

In fact, a critical finding of these studies is the dissociation between sodium balance and arterial pressure. Sodium excretion has consistently been linked to blood pressure regulation in the past (31). In addition, an inability to excrete a chronic sodium load has been proposed to be an important mechanism in nearly all forms of experimental hypertension (12). The fact that these APx animals remained normotensive in light of impaired renal sodium excretion (on a high-salt diet) challenges this idea as a sole mechanism of experimental hypertension. Indeed, these results will lead to further experimentation into the mechanisms of maintaining normal arterial pressure in APx rats despite impaired sodium excretion. These future studies may provide new insights into overall blood pressure regulation and the pathogenesis of salt-dependent hypertension.

Perspectives

In summary, we presently report the maintenance of normal arterial pressure in APx rats when chronically placed on either high- or low-salt diets. Furthermore, APx rats showed no changes in arterial pressure from sham-operated control rats throughout these changes in dietary salt intake. These results do not support the hypothesis that an intact AP is necessary for the maintenance of normotension during chronic changes in dietary salt intake. We feel this is an important finding in our present overall understanding of this CVO and its relation to chronic cardiovascular regulation. Despite these negative findings, we do point out the fact that APx animals had an impaired renal sodium excretory ability when consuming high salt compared with sham control rats. Independent of normal long-term arterial pressure regulation, these results do suggest that the AP plays a role in the long-term neurogenic control of renal sodium handling and excretion during chronic changes in sodium intake.

This study was supported by National Heart, Lung, and Blood Institute Grant HL-50371.


Received 24 April 1998; accepted in final form 8 July 1998.

REFERENCES


