Studies on the Role of the Pituitary in the Control of Atrial Natriuretic Factor Secretion and Sodium Excretion (43084)

JOHN R. DIETZ AND STANLEY J. NAZIAN
Department of Physiology and Biophysics, University of South Florida, College of Medicine, Tampa, Florida 33612

Abstract. **Recent work suggests that hypophysectomized (HYPOX) rats show low levels of atrial natriuretic factor (ANF) and an attenuated diuresis and natriuresis to blood volume expansion. The purpose of this was (i) to examine the effect of various hormone replacements on ANF and renal excretion in HYPOX rats and (ii) to compare the renal responses to exogenous ANF in intact and HYPOX rats. Groups of rats received subcutaneous pellet implant of either dexamethasone (DEX), thyroxine (T_4), or a placebo. Approximately 1 week later, they were anesthetized and subjected to a 20% blood volume expansion. DEX rats had a higher mean arterial pressure than placebo-treated rats while both MAP and heart rate were higher in T_4 rats. Only the DEX rat showed augmented renal responses to volume expansion while no group showed significant changes in plasma ANF concentration during volume expansion. In a second series, groups of HYPOX rats received renal capsular transplants of either six hemi-pituitaries or six pieces of muscle which markedly raised serum prolactin levels in the hemi-pituitary group. The hemi-pituitary rats showed a greater diuresis and natriuresis during volume expansion than the muscle group and also showed a transient increase in plasma ANF. In addition, groups of either intact or HYPOX rats were anesthetized and received intravenous bolus injections of ANF. Both intact and HYPOX rats showed a very similar diuresis and natriuresis to exogenous ANF. However, potassium excretion was markedly reduced in HYPOX rats. The results show that DEX augments the renal responses to volume expansion by some mechanism which does not involve changes in plasma ANF. Thyroxine increases mean arterial pressure and heart rate in HYPOX rats but does not augment the renal or ANF responses to volume expansion. Chronic elevations in prolactin increase the renal response to volume expansion. Finally, the kidneys of HYPOX rats are capable of increasing sodium and water output in response to large doses of exogenous ANF.** [P.S.E.B.M. 1990, Vol 194]

Atrial natriuretic factor (ANF) is a hormone secreted by the heart which increases renal sodium excretion (1). The elaboration of ANF from the heart appears to be important for the normal renal responses to acute volume expansion (2–5). Atrial stretch is a primary determinant of ANF release from the heart (6, 7), although other factors such as changes in heart rate (8–10) may also be important. Both angiotensin II and antidiuretic hormone have been shown to increase plasma ANF levels *in vivo*, but their effects appear to be primarily indirect since they have no direct effects on ANF release *in vitro* when cardiac preload and afterload are controlled (11).

Recently, Zamir *et al.* (12) and Arjamaa *et al.* (13) demonstrated that plasma ANF levels are low in hypophysectomized rats and that hypophysectomized rats show a blunted rise in plasma ANF in response to blood volume expansion (12). Our laboratory confirmed their findings and also showed that hypophysectomized rats display an attenuated diuresis and natriuresis to blood volume expansion (14). The blunted renal response to volume expansion correlated with the reduced levels of ANF. In addition, in experiments using an isolated rat heart-lung preparation, hypophysectomized rats release less ANF than normal rats even under conditions where venous return and aortic pressure were identical in the two groups (14). These findings suggest that one or more pituitary factors may be required for normal ANF secretion and normal renal responses to volume expan-

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sion. Several studies have suggested a role for thyroid hormones in regulating cardiac ANF and ANF secretion (15–21). Glucocorticoids, which are low in hypophysectomized rats, may affect both cardiac and plasma levels of ANF (22, 23). In addition, there is evidence to suggest that prolactin may be necessary for normal ANF secretion (12).

One purpose of this study was to examine the effects of various hormone replacements on ANF and renal function in hypophysectomized rats. A second purpose was to compare the effects of exogenous ANF on renal sodium and water excretion in normal and hypophysectomized rats.

Materials and Methods

Intact and hypophysectomized male Sprague-Dawley rats were purchased from Harlan Laboratories. At the end of the volume expansion experiments described below, the sella turcica of hypophysectomized rats was examined and animals having visible remnants of pituitary tissue were eliminated from further study.

Pellet Implants (Series 1). Hypophysectomized rats were anesthetized with halothane (5–6 days after hypophysectomy) and time release pellets (Innovative Research of America, Toledo, OH) were implanted subcutaneously in the midscapular region. The animals received either dexamethasone (5 mg), thyroxine (5 mg), or a placebo pellet containing only the carrier for the drugs. Eight rats were used per group. These rats were allowed to recover and 7–12 days later were acutely volume expanded as described below. The time release pellets are capable of producing sustained drug levels for up to 21 days.

Others have shown that the thyroxine (T_4) pellets used here will produce constant plasma T_4 levels three to five times normal for at least 2 weeks when implanted in the fashion we have described (24).

Pituitary Implants (Series 2, Hyperprolactinemia). Normal Sprague-Dawley rats were used as donors for the pituitary tissues. They were anesthetized with sodium pentobarbital (50 mg/kg) and decapitated. The pituitaries were removed and placed in buffered saline. The posterior pituitaries were removed and discarded and the anterior pituitaries divided in half.

Hypophysectomized rats were anesthetized with halothane and the left kidney exposed via a flank incision. Either six hemi-pituitaries (pituitary implant, n = 7) or six small pieces of abdominal muscle (control, n = 8) were implanted under the left kidney capsule. The rats that received the transplants were allowed to recover and were used in the volume expansion protocol 7–12 days later as described below. At the end of the acute volume expansion experiments, an additional blood sample was drawn for measurement of serum prolactin by radioimmunoassay as described previously by Nazian (25).

Blood Volume Expansion Experiments. The rats were anesthetized with sodium pentobarbital (50 mg/kg) with supplemental anesthetic being given as needed. The animals were placed on a heating pad and a tracheostomy was performed. Polyethylene catheters (PE-50) were placed in the right carotid artery and right jugular vein, and both ureters were also catheterized (PE-10). The rats were prehydrated with 2 ml of 0.9% saline and allowed 30 min to stabilize. Arterial blood pressure and heart rate were measured from the carotid catheter. Urine was collected in preweighed tubes for four 20-min periods (80 min total). The first 20-min period served as a control (0–20 min). Both Series 1 and Series 2 hypophysectomized rats received a blood volume expansion during the second 20-min urine collection period. The volume of blood infused was 20% of estimated blood volume and was infused over the course of the entire 20-min period (20–40 min). Blood volume was estimated as 6% of body weight. Two additional 20-min urine collections were made after the volume expansion (40–80 min). Blood samples of 2 ml each were drawn from the carotid catheter 5 min before the volume expansion, 5 min after the volume expansion, and at the end of the final urine collection or at 15, 45, and 80 min from the start of the experiment, respectively. The first two blood samples were simultaneously replaced by an equal volume of donor blood given intravenously. All blood samples were placed immediately in prechilled tubes containing EDTA. Plasma obtained after centrifugation was frozen at −20°C until extraction and assay.

Our previous work (14) showed that this degree of blood volume expansion produces a striking increase in plasma ANF concentration in intact rats but little response in hypophysectomized rats.

Source of Donor Blood. Donor blood for blood volume expansions and blood sample replacement was obtained from normal rats. One animal was anesthetized per experiment using halothane and 7–8 ml of blood drawn by cardiac puncture. In a previous study we found that blood sample replacement in this fashion maintained arterial pressure relatively constant and had no significant effect on plasma ANF concentration in a time control series of experiments (14).

ANF Injection (Series 3). The purpose of this series was to determine whether hypophysectomized rats show normal increases in urine flow and sodium excretion to response to exogenous ANF.

Intact and hypophysectomized rats (eight rats per group) were anesthetized, tracheotomized, and prepared with jugular, carotid, and ureteral catheters as described above. Following a 30-min equilibration period, urine was collected for four-min periods. Synthetic ANF (rat ANF 99-126; Sigma Chemical Co., St. Louis, MO) was injected intravenously after the first

(0.1 μg/kg) and third (1.0 μg/kg) urine collection periods.

Extraction of ANF from Plasma. Samples of rat plasma (0.5 ml) were placed on Amprep C_8 cartridges (Amersham) activated with 2 ml of methanol and 2 ml of 0.1 *M* acetic acid. The cartridges were washed with 2 ml of 0.1 *M* acetic acid and the ANF fraction eluted with 1.5 ml of acetonitril/0.1 *M* trifluoroacetic acid, 60:40 (v/v). The eluted sample was evaporated to dryness under a stream of nitrogen gas. Using ^{125}I-labeled ANF, the recovery of ANF by this method was 84 ± 2%. For assay the samples were resuspended in 0.9 ml of assay buffer, centrifuged, and two 300-μl aliquots of the supernatant assayed as described below.

The radioimmunoassay for ANF (26) has been described previously (14, 27) and was used here with minor modifications. The assay employed human ANF 99-126 as a standard (Sigma Chemical Co., St. Louis, MO), rabbit antisera (RAS8798; Peninsula Laboratories, Belmont, CA), and ^{125}I-labeled human ANF as a tracer (IM-187; Amersham Corp., Arlington Heights, IL). The primary antibody was prepared in 0.1% Triton X-100 and 1.4% normal rabbit serum. Two-hundred microliters of a 1/60 dilution of second antibody (Antibodies, Inc.) were added 24 hr after the tracer. After an additional 24 hr, 3 ml of assay buffer were added and the precipitates were pelleted by centrifugation at 3000*g* for 30 min. The supernatants were removed by aspiration. The antibody employed here cross-reacts 100% with both the human and rat forms of ANF.

Urine sodium and potassium concentrations were measured by flame photometry (model 943; Instrumentation Laboratories, Lexington, MA) and urine osmolality was measured by the freezing point depression method (micro-osmette; Precision Systems, Sudbury, MA). All urine flow and electrolyte excretion data were normalized relative to body weight to account for the differences in body weight between the various treatment groups. In the Series 2 rats, left and right kidney urine samples were collected and analyzed separately to determine if either the pituitary or muscle implants had a significant effect on the function of the left kidney. However, since there were no significant differences in left and right renal excretion in this data, the data were combined and presented as in Series 1.

Statistical comparisons between groups were made with an unpaired *t* test in Series 2 and Series 3 and with an analysis of variance in Series 1. Comparisons within the groups were made with an analysis of variance and Fisher's least significant difference test as a post hoc test. In addition, plasma ANF and sodium excretion in Series 2 rats were subjected to regression analysis to obtain a correlation coefficient. In all cases a $P < 0.05$ was considered to be the criterion for statistical significance.

Results

Series 1: Pellet Implants. The results of experiments using pellet implants of either dexamethasone (DEX), T_4, or a placebo pellet in hypophysectomized rats are shown in Figures 1–3. Mean arterial pressure (MAP, Fig. 1, top) was significantly higher in the DEX- and T_4-treated rats compared with the placebo-treated rats ($P < 0.05$). MAP tended to decrease during the experiment and was significantly reduced during the last period in the T_4- and placebo-treated hypophysectomized rats ($P < 0.05$). MAP did not change significantly in the DEX-treated group. Heart rate (Fig. 1, middle) did not change significantly within any group

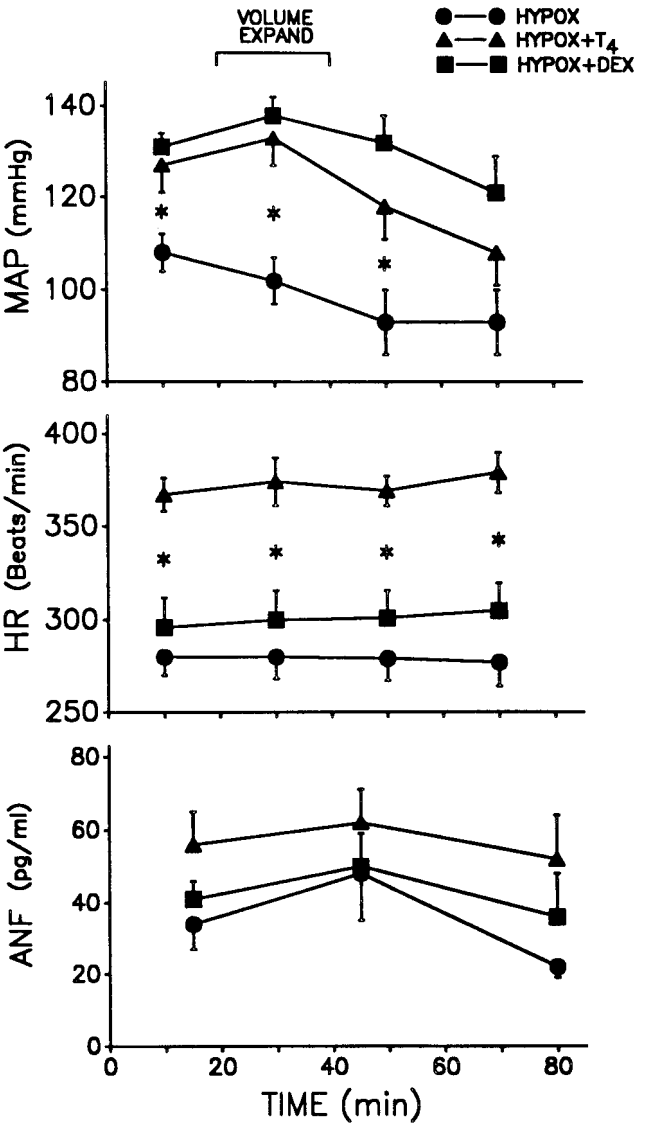

Figure 1. The effect of a 20% blood volume expansion on MAP, heart rate (HR), and the plasma concentration of ANF in anesthetized, HYPOX rats. The groups are placebo-treated (circles), T_4-treated (triangles), or DEX-treated (squares) with eight animals in each group. The volume expansion was performed between 20 and 40 min. Values are mean ± SE. * denotes significant differences ($P < 0.05$) among the groups. Significant differences within groups are noted in the text.

but was approximately 60–80 beats/min higher in the T_4-treated rats throughout the experiment compared with the other two groups ($P < 0.001$).

Plasma ANF concentration (Fig. 1, bottom) tended to increase during the blood volume expansion and to decrease at the last measurement. However, these changes were small and not statistically significant. Although plasma ANF tended to be higher in the T_4-treated rats, there were no significant differences among the groups.

Urine flow (Fig. 2, top) was increased by volume expansion in all three groups ($P < 0.05$). However, urine flow was significantly greater in the DEX-treated group compared with the other group in all but the baseline urine collection period ($P < 0.001$). Sodium excretion (Fig. 2, bottom) increased significantly during volume expansion in both the T_4- and DEX-treated groups ($P < 0.05$, Periods 2 and 3 versus Period 1) but not in the placebo group. Sodium excretion was significantly greater in the DEX-treated group compared with the other two groups during all periods including the control (prevolume expansion) period (all $P < 0.01$). Potassium excretion and total osmolar excretion (Fig. 3) tended to increase in all three groups but increased to a greater extent in the DEX-treated group ($P < 0.05$). Body weight averaged 237 ± 9 g in the placebo-treated hypophysectomized rats and 233 ± 5 g in the T_4-treated

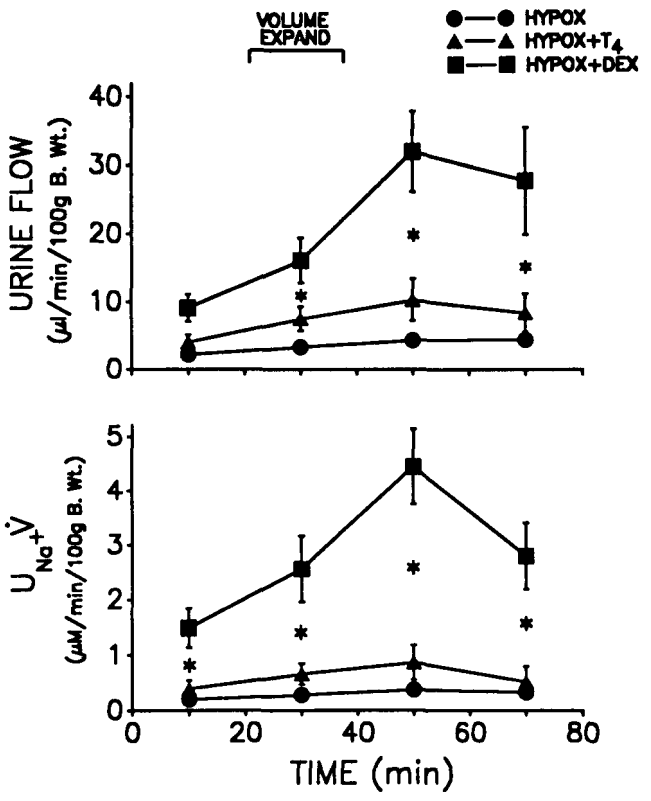

Figure 2. The effect of a 20% blood volume expansion on urine flow and the rate of excretion of sodium ($U_{Na^+}\dot{V}$) which were both factored by body weight. Other descriptions are as indicated in Figure 1.

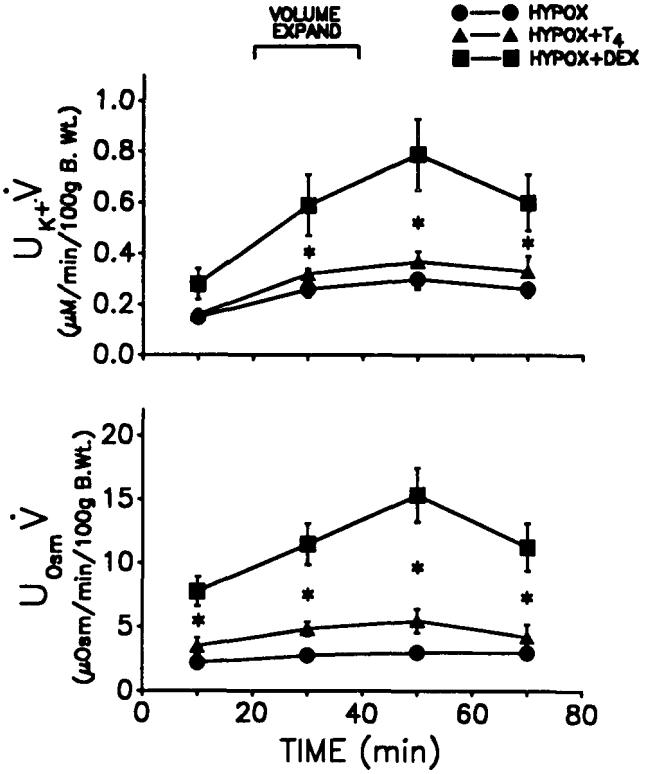

Figure 3. The effect of a 20% blood volume expansion on the rate of excretion of potassium ($U_{K^+}\dot{V}$) and total salutes ($U_{osm}\dot{V}$) which are factored by body weight. Other descriptions are as in Figure 1.

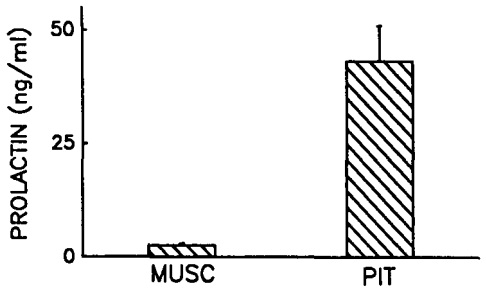

Figure 4. *Serum prolactin levels in anesthetized, hypophysectomized rats 7–12 days after kidney capsule implants of either six hemi-pituitaries (PIT, $n = 7$) or six pieces of abdominal muscle (MUSC, $n = 8$). Values are mean ± SE.*

hypophysectomized rats ($P > 0.05$). Body weight was significantly lower (176 ± 5 g) in the DEX-treated hypophysectomized rats ($P < 0.001$).

Series 2: Pituitary/Muscle Implants. Serum prolactin (Fig 4) averaged 2.6 ± 0.4 ng/ml in hypophysectomized rats which received renal capsule implants of muscle and was significantly greater in hypophysectomized rats which received the pituitary implants (43.2 ± 7.8 ng/ml, $P < 0.001$). Mean arterial pressure and heart rate (not shown) were not different in these two groups of hypophysectomized rats and did not change significantly during the acute volume expansion experiments. Figure 5 shows the renal and ANF responses to the 20% blood volume expansion which was performed

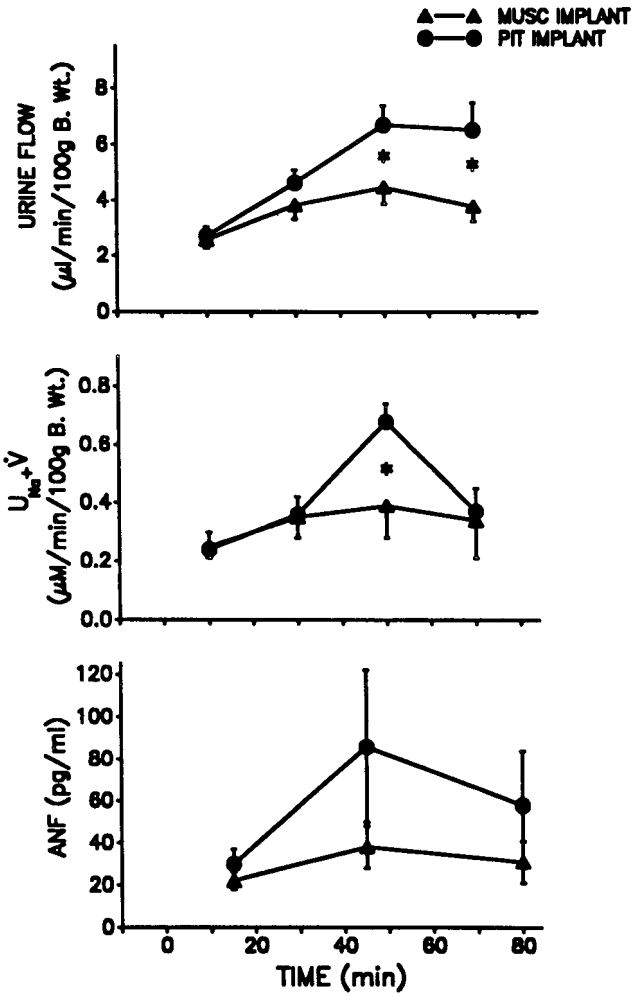

Figure 5. The effect of 20% blood volume expansion on urine flow, the rate of excretion of sodium ($U_{Na+}\dot{V}$), and plasma ANF in anesthetized, hypophysectomized rats. Volume expansion occurred between 20 and 40 min. The rats received kidney capsule implants of either six hemi-pituitaries (circles, PIT, $n = 7$) or six pieces of abdominal muscle (triangles, MUSC, $n = 8$). Values are mean ± SE. * denotes significant differences ($P < 0.05$) between groups. Significant differences within groups are noted in the text.

between 20 and 40 min of the experiments. Volume expansion produced a significantly greater diuresis (upper panel) and natriuresis (middle panel) in hypophysectomized rats that received pituitary implants compared with hypophysectomized rats receiving muscle implants (both $P < 0.05$). Urine output was significantly greater during both collection periods following volume expansion ($P < 0.05$) while sodium excretion only increased transiently (40–60 min, $P < 0.05$). Plasma ANF concentration (Fig. 5, lower panel) tended to increase immediately following volume expansion, but the change was transient and did not quite achieve statistical significance ($F = 2.97$, $P = 0.0744$). However, there was a significant correlation between changes in plasma ANF and changes in sodium excretion in the pituitary implant group ($r = 0.76$, $P < 0.05$).

Series 3: ANF Injections. Mean arterial pressure (Fig. 6, top) was significantly greater in intact rats than hypophysectomized rats prior to the ANF injections ($P < 0.05$). Arterial pressure fell with each injection, with a maximum change from 124 ± 7 to 89 ± 3 mm Hg in the intact rats and from 101 ± 5 to 80 ± 4 mm Hg in the hypophysectomized rats. Urine flow (Fig. 6, middle) and the rate of excretion of sodium (Fig. 6, bottom) both showed small but consistent changes ($P < 0.05$) with the lower dose of ANF (0.1 µg/kg) and marked increases ($P < 0.001$) with the higher dose (1.0 µg/kg) of ANF. However, there were no significant differences in urine flow and sodium excretion between the two groups. Potassium excretion (Fig. 7, top) was significantly lower in hypophysectomized rats compared with intact rats and failed to respond significantly to the bolus ANF injections. Total osmolar excretion (Fig. 7, bottom) was also significantly lower ($P < 0.05$) in hypophysectomized rats during the second urine collection period.

Discussion

In this study, treatment of hypophysectomized rats with dexamethasone (Fig. 2) and chronic hyperprolac-

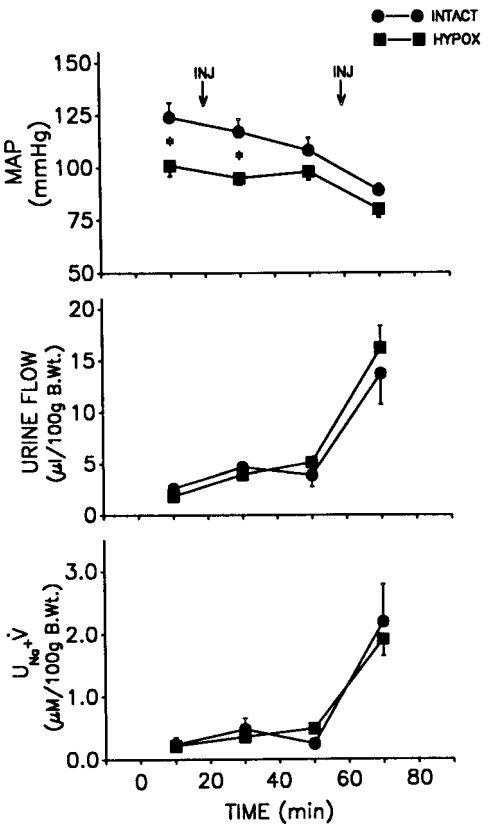

Figure 6. The effect of intravenous bolus injection of ANF on MAP, urine flow, and sodium excretion ($U_{Na+}\dot{V}$) in anesthetized rats, either intact (circles, $n = 8$) or HYPOX (squares, $n = 8$). ANF injections were given at the arrows. The first injection was 0.1 µg/kg body wt, and the second 1.0 µg/kg body wt. Values are mean ± SE. * denotes significant ($P < 0.05$) differences among the groups. Differences within the groups are described in the text.

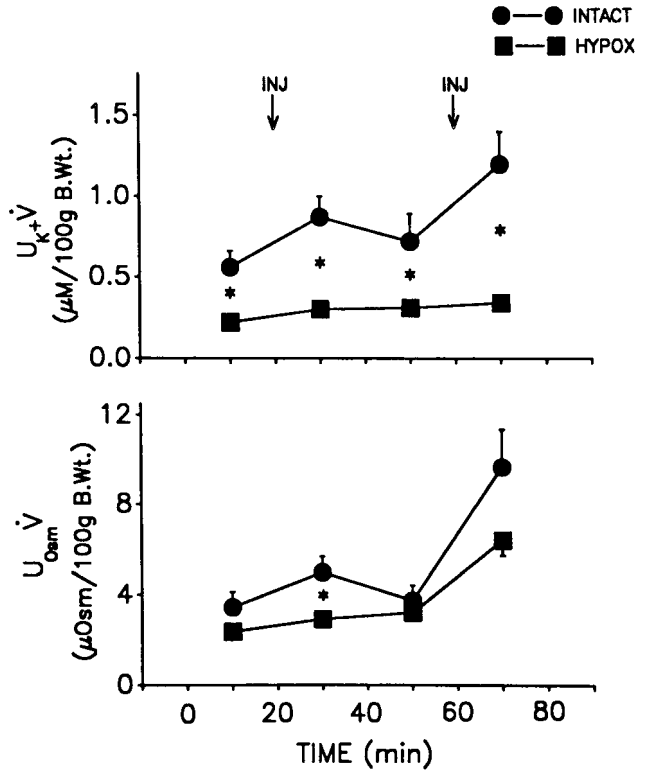

Figure 7. The effect of intravenous ANF injections on potassium and total solute excretion which were factored by body weight. Other descriptions are as in Figure 6.

tinemia (Fig. 5) increased the renal response to blood volume expansion while T_4 treatment failed to produce a significant effect.

In the case of the dexamethasone, at least part of this response could be attributed to the higher arterial pressure and, therefore, renal perfusion pressure in this group. However, hypophysectomized rats treated with T_4 also showed a higher arterial pressure than placebo-treated rats and still failed to show a significantly greater diuresis or natriuresis to volume expansion (Fig. 2). Chronic treatment with glucocorticoids in rats can increase glomerular filtration rate (28), and this could explain the augmented renal responses to volume expansion in the dexamethasone-treated group. Gardner *et al.* (23) treated normal rats with dexamethasone (1 mg/day) and found an increase in ANF mRNA and plasma ANF after 48 hr of exposure to the drug. In the present study, we failed to show a significant effect of dexamethasone on plasma ANF. This difference may be explained by the fact that lower doses of the drug were used here over a longer time interval. Also, our rats were hypophysectomized and the blood samples were drawn in the anesthetized state.

A major point here is that dexamethasone treatment increased the renal responses to blood volume expansion without altering basal plasma ANF concentration of the ANF response to volume expansion. In a previous study, we found an attenuation of both the renal and ANF responses to volume expansion in hypophysectomized rats compared with intact control rats. Changes in plasma ANF and changes in sodium excretion were significantly correlated in these two groups. It is well known that ANF is secreted in response to atrial distention (6) and plasma ANF levels increase during volume expansion in intact animals (7). Several lines of evidence suggest that the natriuresis which occurs in response to volume expansion is dependent on the release of ANF from the heart. Atrial appendectomy attenuates both the rise in plasma ANF (3) and the natriuresis (2, 3) in response to volume expansion in the rat. Benjamin *et al.* (29) found a similar attenuation in conscious atrial appendectomized monkeys with volume expansion. Second, the renal responses to volume expansion can be inhibited by antibodies to ANF (4, 5). In contrast, some recent work suggests that changes in plasma ANF and increases in sodium excretion can be dissociated during atrial distension in the dog (30) or with acute volume expansion in the rat (31). The present results would appear to support this view since dexamethasone treatment augmented the renal responses to volume expansion (Fig. 2) by some mechanism which clearly does not require significant increases in plasma ANF concentration (Fig. 1). This does not rule out an important role for ANF in volume expansion-induced natriuresis. Evidence suggests that dexamethasone can increase ANF receptors in some tissues (32) which could account, at least in part, for the enhanced diuresis and natriuresis in the present study. In addition, volume expansion increases the responsiveness of the kidneys to exogenous ANF (33).

The role of the thyroid in controlling ANF secretion is somewhat controversial. Some workers have shown that ANF is reduced in hypothyroidism and elevated in hyperthyroidism in both rats and humans (16, 20, 21). Also, T_4 treatment in hypothyroid humans has been shown to increase plasma ANF levels (20, 21). In contrast, Ladenson *et al.* (18) found that plasma ANF levels were similar in hyperthyroid, euthyroid, and hypothyroid subjects and plasma ANF was not altered by appropriate treatments in either the hyperthyroid or hypothyroid group. They also found similar plasma ANF levels in hypothyroid rats compared with those of euthyroid rats (18). Obviously, untreated hypophysectomized rats will be hypothyroid due to reductions in plasma thyroid-stimulating hormone levels which could explain the relatively low heart rates in the untreated rats in the present study (Fig. 1). As would be expected, the hypophysectomized rats treated with T_4 showed significantly higher heart rates compared with placebo-treated hypophysectomized rats. Plasma ANF levels tended to be higher in the T_4-treated group, although not significantly and T_4-treated hypophysectomized rats responded to volume expansion similarly

to placebo-treated hypophysectomized rats (Figs. 2 and 3). Therefore, our data suggest that the reduced level of T_4 per se or the reduced heart rate cannot explain the reduced renal and ANF responses observed during volume expansion in hypophysectomized rats.

Several studies have shown that ANF inhibits the release of prolactin from the pituitary (34–37) which suggests a possible negative feedback loop operating between these two hormone systems. In addition, hypophysectomized rats receiving pituitary transplants under the kidney capsule were shown to have elevated ANF levels in one study (12). In the present study, hypophysectomized rats receiving six anterior pituitaries halves under the kidney capsule showed marked elevations in prolactin compared with control hypophysectomized rats (Fig. 4) and an augmented diuretic and natiruretic response to volume expansion (Fig. 5). Although, plasma ANF tended to increase more in the pituitary implanted hypophysectomized rats, the increase was very transient as was the natriuresis in this group. These results suggest that some factor from the anterior pituitary may play a role in the release of ANF from the heart and in the normal renal responses to volume expansion.

Zamir *et al.* (12) also found that anterior pituitaries transplanted to the kidney capsule increased plasma ANF levels in hypophysectomized rats, although, they did not measure renal excretion. They also found that the anterior pituitary transplants had no significant effect on mean arterial pressure. Their study (12) suggested that some hormone or factor other than prolactin mediates the augmentation of ANF release since hypophysectomized rats chronically treated with prolactin failed to show an increase in plasma ANF. In the present study, it is clear that rats receiving pituitary transplants show augmented renal responses to volume expansion (Fig. 5). This augmentation of renal excretion may be mediated by increased secretion of ANF or, alternatively, prolactin may alter kidney function directly by increasing glomerular filtration rate (38) or by altering electrolyte reabsorption (39). In addition, residual secretion of ACTH, thyroid-stimulating hormone, or other factors from the transplanted pituitary may augment renal salt and water excretion by increasing the plasma level of glucocorticoids even though the secretion of ACTH and thyroid-stimulating hormone from the transplants has been shown to be much lower than normal (40).

The final question addressed in the present study concerned the ability of the kidneys of hypophysectomized rats to increase excretion of salts and water in response to exogenous ANF. We predicted that since hypophysectomized rats have a reduced arterial pressure, the rats would show an attenuated diuresis and natriuresis to exogenous ANF. Studies by others indicate that the renal response to ANF is at least partially dependent on renal perfusion pressure (41). Interestingly, hypophysectomized rats showed a similar diuresis and natriuresis to intact rats despite their lower arterial pressure (Fig. 6). However, the hypophysectomized rats did show a markedly attenuated kaliuretic response to the intravenous ANF injections. This attenuated kaliuresis in hypophysectomized rats may possibly reflect reduced levels of aldosterone which is necessary to excrete a potassium load (42). It is possible that chronic reductions in arginine vasopressin could potentially alter renal function, although arginine vasopressin (or lack of) does not appear to play a major role in ANF secretion (43, 44). Regardless, the results do suggest that failure of hypophysectomized rats to increase sodium and water excretion in response to volume expansion cannot be completely attributed to an inability of the kidneys to increase salt and water output.

Although the completeness of hypophysectomy was verified at the end of each of the experiments, it is possible that some small patches of residual pituitary tissue could remain. If so, then these cells could hypersecrete prolactin and this condition could effect ANF secretion. However, in all of the animals where prolactin was measured (Fig. 4), serum prolactin levels were near the lower limit of detection of the assay. Also, none of the hypophysectomized rats grew at normal or near normal rates.

Previous work from this laboratory showed that hypophysectomy decreases ANF secretion and reduces the normal diuretic and natriuretic response to acute blood volume expansion (14). Obviously, removal of the pituitary results in a host of pathophysiologic changes which could contribute to these reduced responses, including lower blood pressure, blood volume, heart rate, basal metabolic rate, growth hormone, and many other factors as well as low plasma ANF. In the present study, we attempted some single hormone treatments in hypophysectomized rats in order to obtain a better understanding of the relationship between ANF and kidney function. None of the single hormone treatments used in the present study were, when used alone, capable of completely restoring all of the renal and hormonal responses to volume expansion. Perhaps future experiments using a combination of treatments will help provide a better understanding of the role of the pituitary in the regulation of ANF secretion and sodium excretion.

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