Comparable Effect of Isotonic Infusions on Blood Pressure in the Anephric Rat

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SUMMARY Whether and to what extent sodium chloride infusions elevate blood pressure acutely were examined in conscious, normotensive, and spontaneously hypertensive (SHR) anephric rats. All animals were bilaterally nephrectomized 18 hours before study.

Normotensive Wistar rats, allowed no food or water post nephrectomy (Groups I–IV), received either no infusion (Group I, control) or intravenous infusion of isotonic solutions of either NaCl, mannitol, or dextrose at a rate of 0.018 ml/min for 2 hours. Mean arterial pressure (MAP) measurements were determined directly by arterial catheter for control (C), 1 hour, and 2 hours. Blood pressure was increased above control in all groups at 1 hour and 2 hours (p < 0.05). The increase in MAP with NaCl was similar to that with no infusion or infusion of mannitol or dextrose. Normotensive Wistar rats (Groups V–VI) and SHR (Group VII) were allowed free access to food and water post nephrectomy and received either no infusion (Group V, control) or infusion of isotonic saline at a rate of 0.037 ml/min for 2 hours. MAP was elevated above control in all groups (V–VII) at 1 and 2 hours (p < 0.05). The magnitude of the rise was similar among all groups. Food and water accessibility post nephrectomy did not alter results. In both sets of experiments when saline was infused we were unable to identify any increase in blood pressure greater than control at either infusion rate. In fact, we continued the saline infusion in Group VI, until 100 ml of saline was infused without any elevation in blood pressure above control. We conclude that during the 2 hours of observation neither sodium nor chloride ions exert an independent effect on MAP in normotensive or hypertensive anephric rats when compared to no infusion or isotonic isovolemic infusions of mannitol or dextrose during the same time period. (Hypertension 5: 421–426, 1983)

KEY WORDS • sodium chloride • mannitol • dextrose • Wistar Kyoto rats • spontaneously hypertensive rats • acute infusions

CHRONIC dietary administration of sodium chloride may elevate blood pressure under a variety of specific conditions.1–3 It has been assumed that this action is mediated indirectly because of accompanying fluid retention and the resultant increase in intravascular volume.4 Although the volume theory is widely accepted, other mechanisms such as release of a slow pressor substance have been suggested5,6 as an alternate means by which sodium chloride indirectly induces an elevation in blood pressure.

Although there exists a large body of literature on chronic sodium administration and blood pressure, there are few studies dealing with the effects of acute sodium administration on blood pressure. In 1980, Hatzinikolaou et al.,7 found higher blood pressures in normotensive anephric rats receiving acute infusions of hypertonic saline than in those receiving hypertonic mannitol. The pressor action of hypertonic saline was not completely inhibited by a vasopressin antagonist, and the difference between the blood pressure after vasopressin (AVP) blockade, and control was greater in the saline group. We inferred that the hypertensive effect following hypertonic saline infusion and AVP blockade could have been due to a direct pressor action of sodium chloride, above the osmotic stimulation of vasopressin. If this is correct, then one might expect that the acute infusion of isotonic sodium chloride should result in a greater pressor activity than infusion of other isotonic solutions, which may even lower blood pressure by dilution. Isotonic, rather than hypertonic solutions, have the advantage of not stimulating the osmoreceptors, and thus avoid the pressor effect of the resultant vasopressin release. It should be recognized that such a series of experiments should be able to address the question of whether isotonic saline has a
pressor action acutely, and answer whether the sodium ion plays a physiologically active or passive role. It leaves open the question of whether concentrations of sodium ions above the normal range are pressor.

Therefore, the present study was undertaken to determine whether isotonic sodium chloride acts acutely as a pressor agent by testing its effect in conscious anephric normotensive and spontaneously hypertensive rats. The anephric preparation was selected because acute administration of sodium chloride to animals with kidneys results in rapid natriuresis. Further, since the level of sodium sensed at the macula densa is inversely related to renin-angiotensin activity, a pressor effect might be masked by a concurrent fall in circulating angiotensin II. Since pressor sensitivity to sodium chloride may vary according to the preexisting level of blood pressure, both normotensive and hypertensive rats were studied.

Methods

Male normotensive Wistar rats and Okamoto-Aoki spontaneously hypertensive rats (SHR), weighing between 225–300 g were maintained on normal Purina rat chow. Seven experimental protocols were carried out. For Groups I–VI we had eight normotensive rats in each group and for Group VII had eight SHR. All animals, 18 hours prior to study, underwent bilateral nephrectomy under Na pentobarbital anesthesia, 30 mg/kg. Following nephrectomy, the animals were allowed either no access or free access to food and water as described below. On the day of the experiment, with the rat under ether anesthesia, the femoral artery and vein were cannulated with PE50 tubing. The rats were restrained and allowed to recover from the anesthetic for at least 90 minutes, before we recorded control values and began a standard 2-hour period of experimentation. Mean arterial blood pressure (MAP) and heart rate (HR) were obtained directly from the arterial catheter by means of a Statham Pressure Transducer, Model 23Db, and recorded on a Gould brush recorder (Gould Inc., Saddle Brook, New Jersey). Prior to infusion, solutions were determined to be isotonic by Osmette Osmometer (Precision Systems, Inc., Sudbury, Massachusetts) and delivered through the venous catheter using a Harvard Infusion Pump (Harvard Instruments, Millis, Massachusetts).

At time zero, 18 hours after nephrectomy, control measurements of blood pressure and HR were recorded. The procedure then differed in accordance with each particular series. Groups I–IV had no access to food or water following nephrectomy. Group I served as control (no infusion). Groups II–IV received infusions at a rate of 0.018 ml/min for a period of 2 hours. The total volume infused was 2.2 ml. Group II received 0.9% NaCl solution; Group III received 5% mannitol solution, and Group IV received 5% dextrose solution.

Groups V–VII were allowed free access to food and water following nephrectomy. Group V served as control (no infusion). Groups VI and VII received infusions at a rate of 0.037 ml/min for 2 hours totaling 4.4 ml. Group VI received 0.9% NaCl solution. In Group VI, after the standard 2-hour period, the infusion rate was increased to 1 ml/min, until a total volume of 100 ml had been infused. Group VII, the only group of SHR, was infused with 0.9% NaCl.

Statistical analysis of the data was performed using paired or unpaired Student t test. Significance was accepted at p < 0.05.

Results

Groups I–IV

Mean arterial pressure (MAP) for rats in control Group I and in experimental infusion Groups II–IV, are shown in figure 1. These groups were not allowed access to food or water after nephrectomy. There was a significant elevation in MAP over 2 hours in all of the groups, including Group I, which received no infusion. The average increase in MAP at 2 hours was 7, 13, and 11 mm Hg in Groups I-IV respectively. Groups II–IV received infusions of 0.9% NaCl solution, 5% mannitol solution, and 5% dextrose solution.

Groups V–VII were allowed free access to food and water following nephrectomy. Group V served as control (no infusion). Groups VI and VII received infusions at a rate of 0.037 ml/min for 2 hours totaling 4.4 ml. Group VI received 0.9% NaCl solution. In Group VI, after the standard 2-hour period, the infusion rate was increased to 1 ml/min, until a total volume of 100 ml had been infused. Group VII, the only group of SHR, was infused with 0.9% NaCl.

Statistical analysis of the data was performed using paired or unpaired Student t test. Significance was accepted at p < 0.05.

![Figure 1. Effect of isotonic infusion on mean arterial pressure (MAP) of normotensive anephric rats, at time zero, 1 hour, and 2 hours. Group I served as a time control group; Group II received an infusion of 0.9% NaCl; Group III received an infusion of 5% mannitol; and Group IV received 5% dextrose. Results are expressed as means ± SEM. *Significance at p < 0.05.](http://hyper.ahajournals.org/doi/abs/10.1161/01.HYP.5.4.422?journalCode=hyp)
each group exhibited a significant rise in MAP throughout the experimental period, the magnitude of the rise was similar, indicating that no particular treatment exerted a distinguishable effect on MAP. The effect on HR is shown in figure 2. There was a small yet statistically significant rise in HR in Group I receiving no infusion. No significant changes were seen in the HR of Groups II–IV receiving infusion of the different solutions.

Effects of Accessibility of Food and Water
To explore the effects of dehydration and starvation, we conducted similar experiments in Groups V–VII in which rats were given free access to food and water following nephrectomy. A comparison was made between control rats in Group I, which were not allowed access to food and water after nephrectomy and control Group V, in which rats were given free access to food and water following nephrectomy. We found a significant rise in MAP over 2 hours and the magnitude of the increase was similar (fig. 3). Therefore, food and water accessibility did not alter the blood pressure results. Heart rate did not rise in Group V (table 1). Group V served as a control for Groups VI and VII. Group VI received an infusion of isotonic NaCl at a rate twice that of Group II for 2 hours, totalling 4.4 ml. The effects on MAP and HR are numerically described in table 1. The MAP was significantly elevated at 1 and 2 hours, but did not increase more than that of control Group V. Heart rate was reduced \( p < 0.05 \) at 1 and 2 hours. Following the standard 2-hour period of experimentation, the rate of infusion in Group VI was increased to 1 ml/min, until a total of 100 ml had been infused. The results are shown in figure 4. As described, MAP was significantly above baseline, but not higher than in control Group V following 4.4 ml of saline, and this same increment in blood pressure was sustained until 20 ml of saline was infused. Following this, MAP began to decline steadily, as did HR. Group VII was the only group of SHR used in this study; the effects of infusing isotonic NaCl solution 4.4 ml at 0.037 ml/min for 2 hours are shown in table 1. Like the normotensive animals, in SHR the MAP was elevated significantly by 1 hour and the magnitude of the increase was similar. Heart rate was significantly reduced as in control animals (Group VI). The magnitude of bradycardia in Groups VI and VII was similar \( p > 0.05 \). The tendency of HR to fall was also seen in control Group V, but it did not achieve statistical significance.

Discussion
In this study, there were no differences in the blood pressure response of normotensive anephric rats to isotonic infusions of saline, mannitol, and dextrose or to no infusion. Surprisingly, blood pressure increased gradually and significantly over 2 hours in all animals.
including those not receiving any infusion. This rise in blood pressure following nephrectomy has been reported previously for rats and dogs, although this finding has not been observed consistently. Several explanations for increases in blood pressure following nephrectomy have been suggested, including reductions in tissue compliance and lack of inactivation of an extrarenal pressor mechanism in the rat. It is also possible that an increase in serum osmolarity accompanied by a concomitant elevation in the circulating ADH titer may contribute to postnephrectomy blood pressure elevation; however, these measurements have not been made.

Although studies examining chronic high sodium intake are fairly numerous, the literature has few reports on the acute effects of sodium on blood pressure. The chronic sodium pressor effect has been better expressed in models that restrict or eliminate kidney function. However, as in the present study, investigators found no immediate rise in pressure; rather, they observed an increase of blood pressure after 1 week but not earlier.

However, the pressor action of chronic sodium administration is not always observed. Wistar rats weaned and raised on high salt diets failed to develop hypertension. In another study, subtotal nephrectomy resulted in hypertension, the incidence of which was increased in rats given saline, but the level of hypertension was similar to those not given saline. The studies of Lucas and Floyer and Pelling and Ledingham have shown that nephrectomized rats chronically receiving either 0.5% or 1% saline as drinking water become hypertensive over a period of several days. However, in the Pelling and Ledingham study, allowing the rats to drink a large supplement of water rather than saline also increased blood pressure significantly to the same extent. Thus, it appears that saline administration was not the common denomina-
EFFECT OF ISOTONIC INFUSIONS IN THE ANEPHRIC RAT/Kleinert et al. 425
tor, since sodium-free fluid increased blood pressure. In these chronic studies, saline administration itself did not cause hypertension.

In our acute study, infusion of 0.9% sodium chloride did not induce a pressor response compared to fluid-infused or noninfused controls. Our animals were not allowed to drink during the period of experimentation. Thus, isotonic sodium chloride itself, administered acutely, did not act as a pressor agent in our preparation. We also studied the effect of allowing free access or no access to food and water during the time period between nephrectomy and experimentation, only to find no difference with respect to blood pressure between the groups. Some studies on the effects of hydration and dehydration on blood pressure corroborate our findings, but others disagree. Chronic salt-loading hypertension has been suggested to be a precursor to a resultant volume overload hypertension. In our study, however, the results of Group VI rats that received a saline infusion of 100 ml indicate that acute hypervolemia induced by saline infusion is not in itself a means of eliciting an acute pressor response. MAP continued to decline steeply and steadily as the infusion volume was increased above 40 ml of isotonic saline. It is possible that the cardiovascular system was failing to adequately handle a volume overload of that magnitude, resulting in cardiac failure. It may well be that in our study saline infusion was not carried out long enough to exhaust the venous capacity or, more likely, to eliminate the baroreflex buffer system. Cowley and Guyton demonstrated that the baroreceptor reflex slows the development of salt-loading hypertension by altering total peripheral resistance until a level of steady-state hypertension is developed. It is possible that our experiments were not carried out long enough to see a response.

The Hatzinikolaou et al. study employing acute infusion of hypertonic saline raised the possibility of an independent sodium pressor effect. It is possible that only a partial blockade of vasopressin was achieved in their study. We also considered that they acutely raised the sodium chloride concentration far above the physiological level and that their administering sodium chloride solution to achieve a serum concentration 10% above the normal range may have been vasoconstrictive. A concentration-mediated vasoconstrictive effect of sodium would be unlikely in an acute study such as ours in which isotonic saline was employed. Their study and ours are in agreement concerning the absence of an acute volume effect. On the other hand, it might be postulated that elevations in blood pressure were due to the effect of hypertonic saline drawing intracellular fluid into the circulation. In our experiment (Group VI), we gave a greater total volume of saline and ions but in a lower concentration. Our results show that acute elevations in the number of sodium ions and also volume do not induce hypertension. Thus, we cannot further clarify their results.

Comparison of the isotonic solutions was made in an effort to detect a possible specific acute pressor effect of sodium. The hemodynamic effects of isotonic mannitol and dextrose were indistinguishable from saline. If isotonic sodium chloride exerted a direct independent pressor action in an acute preparation, we would not expect the effects of mannitol and dextrose on blood pressure and HR to be comparable.

Several investigations in humans and rat have presented evidence that predisposition to hypertension prior to sodium chloride loading sensitizes the subject to the pressor effect of sodium. In the present study, a pressor effect could not be demonstrated in the SHR, which were all established to be hypertensive before initiating infusion. Thus, in an acute experiment lasting 2 hours, a pressor effect was not exerted by infusion of sodium chloride, regardless of the baseline level of blood pressure.

Heart rate did not rise in the control group allowed free access to food and water (Group V), suggesting that dehydration may indeed have contributed to the slight increase in HR seen in control Group I, denied access to food and water. Bradycardia occurred in Group VI (normotensive anephric rats) and Group VII (anephric SHR), both of which shared in common free access to food and water following nephrectomy and experimental infusion of 4.4 ml of isotonic saline over a period of 2 hours. It is possible that the larger volume of saline induced a reflex reduction in HR, not observed in Group II, which received only 2.2 ml of isotonic sodium chloride over 2 hours. Reflex bradycardia may be manifested independently of reflex changes in total peripheral resistance. The magnitude of bradycardia experienced by the normotensive rats (Group VI) and the SHR (Group VII) was similar.

Our findings indicate that a sodium chloride or volume overload pressor effect sometimes seen in particular chronic preparations was not demonstrable acutely in the anephric rat in spite of the initial level of blood pressure. From the results of our study, we conclude that: 1) sodium chloride administered in large excess as an isotonic solution does not act directly as an acute pressor agent in normotensive or SHR anephric rats, and thus, does not have an intrinsic pressor property; and 2) isovolemic infusions of isotonic sodium chloride, mannitol, and dextrose do not effect blood pressure and HR in the normotensive conscious anephric rat.

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References

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