Lesions of the Anteroventral Third Ventricle Prevent Salt-Induced Hypertension in the Borderline Hypertensive Rat

Brian J. Sanders and Alan Kim Johnson

Many forms of experimental hypertension depend on the integrity of the periventricular tissue surrounding the anteroventral third ventricle. The current investigation examined the extent to which this forebrain area is necessary for the elaboration of salt-induced hypertension in the borderline hypertensive rat. Eight-week-old male rats were given either electrolytic lesions of the anteroventral portion of the third ventricle region or sham lesions. All rats were then placed on a high salt diet (8% NaCl) for 10 weeks. At the conclusion of this dietary period, direct measurement of resting mean arterial pressure revealed that borderline hypertensive rats with lesions of the anteroventral portion of the third ventricle had significantly lower blood pressure (128.4±5.1 mm Hg) compared with sham-operated rats (148.1±4.1 mm Hg). (Hypertension 1989;14:619-622)

Increases in dietary salt intake have been demonstrated to raise arterial pressure in a variety of animal models, as well as in humans with borderline hypertension. Studies with the spontaneously hypertensive rat (SHR) and the Dahl salt-sensitive model have illustrated the importance of genetic influences in determining the pressor response to a salt challenge. Several recent studies have addressed the issue of salt sensitivity in another genetic model of environmentally induced hypertension. The borderline hypertensive rat (BHR) is the first filial offspring of SHR and the normotensive Wistar-Kyoto (WKY) rat and therefore possesses a predisposition for high arterial pressure. The BHR will not become spontaneously hypertensive like its parent, but increases in dietary salt or long-term exposure to behavioral stress will provoke the expression of hypertension in this model.

Possible mechanisms involved in the pathogenesis of stress- and salt-induced hypertension in the BHR have been previously addressed. These studies have suggested that disruptions in autonomic and renal function may be major contributors to the development of this disorder. However, the participation of central neural structures in the hypertensive process in the BHR has not been investigated until recently. The previous studies demonstrating that ablation of the periventricular tissue surrounding the anteroventral portion of the third ventricle (AV3V) prevents the development of many forms of experimental hypertension led us to recent research showing that this manipulation also inhibits the development of conflict stress-induced hypertension in the BHR.

Because exposure to stress and a high salt diet are considered of potential importance in the production of human essential hypertension, the BHR may represent an applicable animal model with which to study the interaction of environmental factors, genetics, and arterial pressure. Furthermore, the contribution of the AV3V region to the regulation of physiological systems involved in the hypertensive process (namely, autonomic and body fluid balance) suggests a potential role for this area in hypertension in the genetically predisposed BHR. In the current study, we have sought to extend our previous finding that AV3V lesions prevent stress-induced hypertension in the BHR and have investigated whether the expression of hypertension observed in BHR after long-term maintenance on a high salt diet depends on the integrity of the AV3V region.
Materials and Methods

Animals

This study used male BHR. These rats were the first generation offspring of SHR female and WKY male rats purchased from Taconic Farms (German-town, New York). Parent strains were not studied because WKY rats are resistant to the hypertensive effects of a high salt diet and SHR are genetically destined to become hypertensive, thus complicating the analysis of the role of exogenous factors in the disease process. All rats were bred in the animal facility in the Department of Psychology at the University of Iowa. The rats were weaned at 3 weeks of age and individually housed at 7 weeks of age. Standard laboratory rat chow (Purina Chow 5012, Purina Mills, Inc., St. Louis, Missouri) and tap water were available to all subjects until the dietary regimen was instituted as described below.

Experimental Protocol

At 8 weeks of age, all rats were randomly assigned to one of two groups. Indirect blood pressure measurements by the tail-cuff method (IITC, Inc., Woodland Hills, California) revealed no difference in systolic pressure between the two groups (sham lesion, 146.5±1.3 mm Hg vs. AV3V lesion, 145.8±2.26, p>0.80). Subjects in the experimental group received electrolytic ablation of the AV3V region. The rats were anesthetized with a ketamine/acepromazine (5:1 cc) mixture (50 mg/kg i.p.) and positioned in a Kopf stereotaxic instrument. Bregma and lambda were exposed, the head was leveled, and a midline opening was made in the skull 0.5 mm caudal to bregma. A lesioning electrode (24 gauge nichrome wire insulated except at the tip) was lowered on the midline 0.3 mm caudal to bregma to a depth of 7.5 mm from dura. Direct anodal current (2-3 mA) was passed for 25-30 seconds (rectal cathode). Sham-lesion rats were treated similarly except that the electrode was lowered to 4 mm below dura and no current was passed.

After a 2–3-week recovery period, indirect systolic blood pressure measurements revealed no difference between the groups (sham lesion, 146.6±1.3 vs. AV3V lesion, 148.8±0.77, p>0.25). At this time, all rats were placed on a pelleted diet containing 8% sodium chloride (ICN Biochemicals, Cleveland, Ohio) for 10 weeks. This dietary regimen has been previously demonstrated to provoke substantial increases in arterial pressure in the BHR. After the rats had been in a quiescent state for at least 1 hour, baseline cardiovascular measures were taken in all rats before transfer to a Plexiglas footshock chamber (25×30×30 cm). After the rat was placed in the chamber, mild electric shock pulses were administered through the stainless steel grid floor for a 5-minute period (1.0 mA, 0.5 seconds in duration every 5 seconds). Cardiovascular data were recorded after transfer and immediately and 5 minutes after cessation of footshock.

Histology

At the conclusion of the experiment, all subjects were anesthetized as described above and perfused transcardially with 0.9% saline followed by 10% formalin. The brains were removed and stored in the fixative until frozen sections were taken and stained for Nissl substance with cresyl violet.

Statistics

The data collected in this experiment were analyzed with analyses of variance. Analysis of variance with repeated measures (referred to as time) was used to examine the transfer and afterfootshock data. Significant effects were further analyzed with Tukey’s honestly significant difference test. A 0.05 level was chosen for statistical significance. Data are presented as the mean±SEM.
Discussion

The major finding of the current investigation is the demonstration that the integrity of the AV3V is necessary for the development of salt-induced hypertension in the BHR. Many previous studies have illustrated the involvement of the AV3V in the control of the circulation and maintenance of body fluid balance. Given that hypertension provoked by a high salt diet is likely to involve contributions of both of these systems, it is in this context that the prophylactic actions of AV3V lesions may be examined.

One potential mechanism by which ablation of the AV3V region may protect BHR from salt-induced hypertension is by preventing the volume expansion that may attend a high salt diet. Previous investigations have suggested that the ability of the organism to maintain body fluid homeostasis may be important in the development of environmentally induced hypertension in the BHR. For example, BHR maintained on a high salt diet display an antinatriuretic response to acute behavioral stress that is absent in WKY rats, suggesting a possible role for neurally mediated alterations in renal function in the pathogenesis of hypertension in the current model. It is also important to note in this regard that AV3V lesions inhibit the release of vasopressin. Diminution in the availability of this peptide would presumably disrupt the animal’s ability to effect volume expansion. Thus, lesions of the AV3V region may contribute to the prevention of salt-induced hypertension in the BHR by blocking the release of vasopressin and by eliminating a critical central neural influence affecting renal function, both of which would presumably be involved in volume expansion.

Lesions of the AV3V region may also exert an antihypertensive action by reducing the exaggerated autonomic functioning that accompanies salt loading in some animal models of hypertension. For example, the hypertensive progenitor of the BHR responds to a high salt diet with an increase in urinary and plasma norepinephrine and an enhanced depressor response to ganglionic blockade. The latter observation has also been made in the BHR, suggesting an important role for the sympathetic nervous system in the maintenance of salt-induced hypertension in this model. Additionally, the demonstration of descending and ascending neural projections between this forebrain region and other central areas involved in car-

Results

Histological examination verified the lesion placement in the experimental subjects. Damage to the organum vasculosum of the lamina terminalis, median preoptic nucleus, and preoptic-anterior hypothalamic periventricular tissue was observed in all rats in which salt-induced hypertension failed to develop.

Figure 1 (top panel) summarizes the baseline mean arterial pressure of the two groups. Analysis of variance performed on these data revealed a significant main effect of treatment \( (p \leq 0.02) \). Sham-operated BHR had significantly higher mean arterial pressure than BHR with AV3V lesions (148.1±4.1 vs. 128.4±5.1 mm Hg, respectively). Thus, AV3V lesions prevented the development of salt-induced hypertension in BHR and kept blood pressure at a level comparable with BHR maintained on a normal salt diet. The baseline heart rate data are also summarized in Figure 1 (bottom panel). There was no significant difference between groups with respect to this variable (sham lesion, 377±16 vs. AV3V lesion, 344±12.8 beats/min, \( p \geq 0.17 \)).

Figure 2 (top panel) depicts the change in mean arterial pressure to transfer and footshock. As can be seen, although BHR with AV3V lesions had a tendency for somewhat larger changes in pressure, there was no difference in the pattern of response between the two groups \( (p \geq 0.38) \). Changes in heart rate to transfer and footshock are summarized in Figure 2 (bottom panel). Analysis of variance performed on these data indicated a significant group-by-time interaction effect \( (p \leq 0.01) \), indicating that the two groups responded differently to the acute stress paradigm. Subsequent follow-up analyses revealed that at 5 minutes after footshock sham-operated BHR had returned closer to baseline compared with BHR with AV3V lesions (+36±22 vs. +134±8.9 beats/min above baseline, respectively).

FIGURE 2. Line graphs showing changes in mean arterial pressure (top panel) and heart rate (bottom panel) in response to transfer to footshock chamber and immediately (Imm.) and 5 minutes (5') after acute stress in borderline hypertensive rats with sham lesions and with lesions of the anteroventral portion of the third ventricle (AV3V).

Sanders and Johnson

AV3V Lesions and Salt-Induced Hypertension

Downloaded from http://hyper.ahajournals.org/ by guest on October 2, 2017
diovascular regulation, such as nucleus of the solitary tract, is consistent with the notion that the AV3V may contribute to the hypertensive process. BHR with AV3V lesions may be protected from salt-induced hypertension because they lack this important hypothalamic contribution, which would increase peripheral sympathetic nervous system activity and elevate arterial pressure.

We have recently reported that lesions of the AV3V region are also capable of preventing the development of conflict stress-induced hypertension in the BHR. These data coupled with the current observations suggest a provocative similarity between these two forms of environmentally induced hypertension. That is, previous evidence suggests that hypertension provoked by exposure to stress may be mediated in large part by an exaggerated responsiveness of the sympathetic nervous system. On the other hand, increases in arterial pressure resulting from maintenance on a high salt diet are widely thought to be intimately related to volume expansion. Thus, although these two forms of hypertension may be characterized by perturbations in different peripheral systems, these two studies suggest that the AV3V region is a common neural structure underlying both forms of the disorder. Finally, the observation that lesions of the AV3V region prevent these two forms of hypertension in the BHR but are ineffective at interdicting hypertension of the SHR parent suggests a certain degree of independence in the mechanisms mediating the disorder in the two strains. For example, it is possible that AV3V lesions exert their antihypertensive action in BHR by interfering with physiological systems that are inactive in SHR or unaffected by the lesion.

The cardiovascular responses to the acute stress paradigm in this study are reminiscent of previous investigations. For example, one recent study of the effects of AV3V lesions on stress-induced hypertension in BHR revealed that lesioned rats were still capable of transiently raising arterial pressure. Additionally, BHR made hypertensive either by exposure to behavioral stress or increased dietary salt intake typically show very little further increase in arterial pressure to an acute episode of stress. A second study of AV3V lesions in BHR also showed that lesioned rats were still capable of transiently raising arterial pressure. The present study has illustrated the importance of the AV3V in the expression of hypertension induced by a high salt diet in an animal model with a genetic predisposition for the disorder. Perturbations in renal and autonomic functioning have previously been implicated in stress- and salt-induced hypertension in the BHR and have therefore provided potential candidate mechanisms by which AV3V lesions are protective. The present results invite further investigations to examine the extent to which additional central and peripheral systems may be involved in the antihypertensive effects of AV3V lesions.

References

7. Sanders BJ, Cox RH, Lawler JE: Cardiovascular responses to the acute stress paradigm in this study are reminiscent of previous investigations. For example, one recent study of the effects of AV3V lesions on stress-induced hypertension in BHR revealed that lesioned rats were still capable of transiently raising arterial pressure. Additionally, BHR made hypertensive either by exposure to behavioral stress or increased dietary salt intake typically show very little further increase in arterial pressure to an acute episode of stress. A second study of AV3V lesions in BHR also showed that lesioned rats were still capable of transiently raising arterial pressure. The present study has illustrated the importance of the AV3V in the expression of hypertension induced by a high salt diet in an animal model with a genetic predisposition for the disorder. Perturbations in renal and autonomic functioning have previously been implicated in stress- and salt-induced hypertension in the BHR and have therefore provided potential candidate mechanisms by which AV3V lesions are protective. The present results invite further investigations to examine the extent to which additional central and peripheral systems may be involved in the antihypertensive effects of AV3V lesions.

KEY WORDS: anteroventral third ventricle; salt-induced hypertension; borderline hypertensive rats
Lesions of the anteroventral third ventricle prevent salt-induced hypertension in the borderline hypertensive rat.
B J Sanders and A K Johnson

Hypertension. 1989;14:619-622
doi: 10.1161/01.HYP.14.6.619

Hypertension is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1989 American Heart Association, Inc. All rights reserved.
Print ISSN: 0194-911X. Online ISSN: 1524-4563

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://hyper.ahajournals.org/content/14/6/619

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Hypertension can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Hypertension is online at:
http://hyper.ahajournals.org/subscriptions/