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Renal Denervation Eliminates the Renal Response to Continuous Positive-Pressure Ventilation (40599)[1]

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Elevation of the expiratory pressure during mechanical ventilation is frequently associated with a decrease in urine flow and electrolyte excretion (1–3). Although major emphasis has been placed on intrathoracic volume receptors and antidiuretic hormone (4, 5), until recently, very little definitive information concerning the mechanism of the reflex antidiuresis associated with continuous positive pressure ventilation (CPPV) has been reported.

In a previous study, we demonstrated that selective denervation of the aortic arch and carotid sinus baroreceptors eliminated the renal response to CPPV using a positive end expiratory pressure (PEEP) of 10 cm H_2O; in contrast, bilateral cervical vagotomy did not alter the antidiuresis or antinatriuresis observed during CPPV (6). This is in accordance with the earlier work of Baratz *et al.* (7) and Tarak and Chaudhury (8) and provides additional evidence that cardiopulmonary receptors, whose afferent nerve fibers are in the vagi, do not initiate this response. Tarak and Chaudhury (8) have also reported that CPPV can cause suppression of urine flow without elevated levels of antidiuretic hormone.

One possible explanation for the arterial baroreceptor mediated antidiuresis and antinatriuresis associated with CPPV may be related to an alteration in renal nerve activity. It is well documented that the carotid sinus and aortic arch baroreceptors participate in controlling renal nerve activity. Kezdi and Geller (9) have demonstrated that an inverse relation exists between carotid sinus pressure and renal sympathetic nervous discharge and that this relation is essentially linear over the physiologic range of pressure. Furthermore, alterations in renal hemodynamics and electrolyte excretion, independent of renal perfusion pressure, have been reported during occlusion of the common carotid artery (10, 11).

In view of the foregoing discussion, we felt that an evaluation of the participation of the renal nerves as the efferent limb of this reflex was warranted. This paper describes studies which were performed to evaluate the role of the renal nerves in mediating the reflex antidiuresis and antinatriuresis observed during CPPV.

Methods. Seven female dogs underwent unilateral renal denervation 4–5 days before the final experiment. Anesthesia was induced by intravenous injection of sodium thiopental (18 mg/kg) and was maintained by connecting the dog to a closed circuit anesthesia system containing 0.5–1.0% methoxyflurane in oxygen and nitrous oxide. The left kidney was then exposed by a transverse left-flank incision. The renal artery and vein were carefully stripped of nerves and adventitia and painted with 8% phenol. The ureter was stripped of its fibrous coat approximately 5 cm proximal to the kidney and all visible nerves were cut. Care was taken to prevent damage to adjacent structures. The dogs were treated postoperatively with penicillin.

On the day of the experiment, the dogs were anesthetized with intravenous sodium pentobarbital (30 mg/kg) and approximately 1 mg/kg additional anesthesic was administered every 15 min to maintain anesthesia. The dogs were intubated with a large bore endotracheal tube and the cuff was inflated to a gas tight fit. Ventilation was controlled using a volume ventilator (Harvard Apparatus Respiration Pump, Model 607) with a tidal volume of 15 ml/kg and a respiratory frequency of 12 breaths per minute and an inspiration:expiration time ratio of 1:1.5. Ar-

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terial blood gases and pH were monitored intermittently by sampling blood from the aorta and analyzing it with a Corning 165 blood gas analyzer to assure normal ventilatory parameters (12). The dogs were paralyzed with succinylcholine (1 mg/kg).

Catheters were placed in the thoracic aorta via the left femoral artery and the inferior vena cava via the left femoral vein. A rigid catheter was placed in line with the endotracheal tube for measurement of proximal airway pressure. Pressures were measured with Statham $P_{23}Db$ transducers and recorded on an Electronics for Medicine DR-8 recorder.

In preliminary experiments, it was observed that there was a tendency for systemic arterial pressure to decrease during CPPV. Because a decrease in systemic arterial pressure can produce an antidiuresis directly by reducing renal perfusion pressure (13, 14), we decided to maintain a constant pressure of 100 mm Hg within the renal arteries throughout the experiment. Renal perfusion pressure was held constant by adjusting the tension on a snare which was placed around the abdominal aorta approximately 3 cm proximal to the right renal artery. Renal perfusion pressure was monitored by a catheter placed via the right femoral artery in the abdominal aorta distal to the renal arteries. The pressures in the aorta both proximal and distal to the snare were measured continuously. The pressure recorded below the snare was considered to represent renal perfusion pressure.

Ringers lactate containing succinylcholine (0.138 mg/ml), inulin (12.50 mg/ml), and *p*-aminohippurate (7.58 mg/ml) was infused intravenously at a rate of 0.06 ml/kg min. A mild diuresis was established by the intravenous infusion of isosmotic mannitol at a rate of 0.03 ml/kg min. Urine was collected at 30-min intervals via ureteral catheters (PE 205) for the separate measurement of the function of the right and left kidneys. At the midpoint of each urine collection a 10-ml blood sample was withdrawn. The volume of blood withdrawn was replaced with an equal amount of 6% dextran in normal saline in which the red blood cells from the previous collection period were resuspended. Plasma samples and urine samples were analyzed for the concentration of sodium, inulin (15), *p*-aminohippurate (16), and osmolality.

After two 30-min control periods in which urine flow remained relatively stable (± 10% of baseline values), the ventilatory pattern was changed from intermittent positive pressure ventilation (IPPV) to CPPV by applying a positive end expiratory pressure (PEEP) of 10 cm H_2O. PEEP was produced by partial static inflation of an occlusive balloon manifold incorporated into the expiratory limb of the ventilator circuit. The PEEP was maintained for 60 min after which an appropriate recovery period of IPPV followed.

Statistical analysis was performed utilizing a one-way analysis of variance for repeated measures on the same factor (17). This was followed by a Dunnett multiple range *t* test to determine which means, at the 0.05 level of significance, were statistically different from the mean of the control period (18). A Student's paired *t* test (19) was used to determine if significant differences in renal function existed between the right and left kidneys during the control periods in order that we might define residual effects of the prior surgery.

Results. A summary of the hemodynamic data in the unilaterally renal denervated dogs is presented in Figure 1. The application of CPPV produced a significant increase in heart rate and a significant decrease in pulse pressure. Although mean arterial pressure decreased slightly, the difference was not statistically significant. Renal perfusion pressure was held constant at 100 mm Hg.

The application of CPPV produced significant decreases in urine flow, sodium excretion, effective renal plasma flow, and glomerular filtration rate in the right kidney with intact renal nerves (Fig. 2). Chronic renal denervation eliminated the reflex renal response observed during CPPV. Elevation of the expiratory pressure produced a significant decrease in osmolar clearance in the right kidney whereas no significant changes were observed in the filtration fraction or free water clearance (Fig. 3). No significant changes in arterial blood gases or pH were observed during CPPV.

During the control periods, urine flow and sodium excretion were increased ($P < 0.05$) in the left denervated kidney when compared to the right kidney with intact renal nerves, whereas no differences were observed in ef-

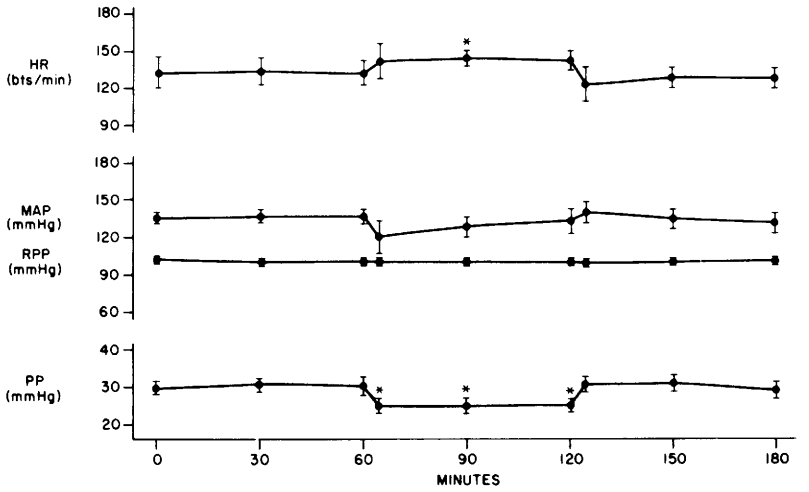

FIG. 1. Effect of continuous positive-pressure ventilation on hemodynamics in the unilaterally renal denervated dogs. Values represent means ±SEM for seven experiments performed on seven dogs. HR = heart rate; MAP = mean arterial pressure; RPP = renal perfusion pressure; PP = pulse pressure. An asterisk above a data point indicates a significant difference, at the 0.05 level of significance, from the mean of the control level.

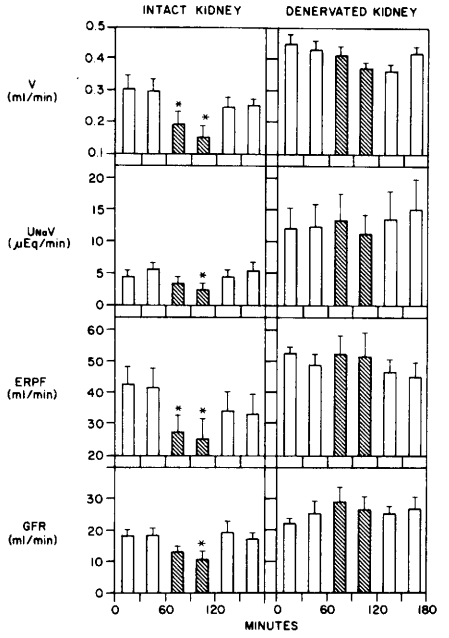

FIG. 2. Effect of continuous positive-pressure ventilation on renal function in the unilaterally renal denervated dogs. Values represent means ±SEM for seven experiments performed on seven dogs. V = urine flow; $U_{Na}V$ = sodium excretion; ERPF = effective renal plasma flow; GFR = glomerular filtration rate. An asterisk above a data point indicates a significant difference, at the 0.05 level of significance, from the mean of the control level.

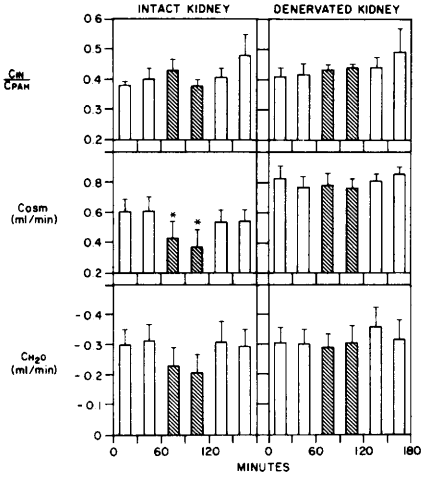

FIG. 3. Effect of continuous positive pressure ventilation on renal function in the unilaterally renal denervated dogs. Values represent means ±SEM for seven experiments performed on seven dogs. C_{IN}/C_{PAH} = filtration fraction; C_{OSM} = osmolar clearance; C_{H_2O} = free water clearance, An asterisk above a data point indicates a significant difference, at the 0.05 level of significance, from the mean of the control period.

fective renal plasma flow or glomerular filtration rate.

Discussion. We have studied the renal response to continuous positive pressure ventilation (CPPV) and have demonstrated that renal denervation eliminates the reflex de-

crease in urine flow (2, 6, 7, 20), sodium excretion (2, 6, 7), effective renal plasma flow (5, 20), and glomerular filtration rate (2, 6, 7, 20) observed during mechanical ventilation with a positive end expiratory pressure. Because both the denervated kidney and the kidney with intact renal nerves were exposed to the same hormones, we feel that changes in circulating levels of antidiuretic hormone and catecholamines are not important in this reflex response.

The results of this study are similar to the earlier reports of Knoefel *et al.* (21) who studied a group of anesthetized dogs during a 40-min period of 9–31 mm Hg continuous positive airway pressure and observed a difference in the renal response by comparing one kidney which had been acutely denervated with the contralateral kidney with intact renal nerves. These investigators utilized an acute denervation model which has been subjected to criticism (22, 23). Katz (23) has demonstrated that some acute denervation models, which employ stripping away hilar nerves and transection of the renal artery, may traumatize the kidney severely and nullify the experimental results. Furthermore, Handley and Moyer (22) demonstrated in the dog and Mercer (24) in the rat, that renal blood flow and glomerular filtration rate may decrease as much as 50% with renal manipulation. Knoefel *et al.* also reported a very heterogeneous renal response to CPPV which they attributed to surgical trauma or the depth of anesthesia. The above consideration, plus the fact that they did not utilize statistics to analyze their data, makes the interpretation of the significance of the results very difficult.

To determine if residual effects of the denervation procedure existed during our experiments, we compared the base-line renal variables of the left denervated kidney to the base-line renal variables of the right intact kidney. There were no differences in effective renal plasma flow or glomerular filtration rate, but there were small differences in urine flow and sodium excretion. We feel that the differences in urine flow and sodium excretion are not residual effects of the prior surgery, but rather, are related to the pentobarbital anesthesia. Pentobarbital anesthesia has been demonstrated to increase renal nerve activity (25), which may in turn decrease urine flow and sodium excretion without altering renal blood flow or glomerular filtration rate (26). Thus, we believe that a recovery period of 4–5 days was adequate.

In our study, the application of 10 cm H_2O positive-end expiratory pressure for 60 min produced decreases of 41, 39, and 32% in urine flow, sodium excretion, and glomerular filtration rate, respectively, in the right kidney with intact renal nerves. These findings are in agreement with the work of other investigators (2, 7). Therefore, we feel that our animals provided a suitable model in which to define the role of the renal nerves in mediating the renal response to continuous positive pressure ventilation.

Tucker and Murray (27) have studied the cardiorespiratory effects of CPPV using a positive-end expiratory pressure of 10 cm H_2O and suggested that the hemodynamic changes which occurred were insufficient to cause renal vasoconstriction. These investigators demonstrated that during CPPV the balance between renal oxygen supply and renal oxygen consumption was constant as evidenced by a constant renal arteriovenous oxygen difference and a constant renal vein P_{O2}. Renal oxygen consumption, however, is not directly dependent on renal blood flow but rather, on the renal transport of sodium (28, 29). Therefore, it is possible that renal blood flow may have decreased in their study without a change in arteriovenous oxygen difference.

Our data suggest that the antidiuresis and antinatriuresis associated with elevation of the expiratory pressure may be attributed to altered renal hemodynamics secondary to increased renal sympathetic vasoconstrictor activity. The fact that we observed a significant decrease in the effective renal plasma flow and in glomerular filtration rate without a significant change in the filtration fraction suggests that the vasoconstriction primarily involves the afferent arteriole. The decrease in osmolar clearance appears to be predominately related to the decrease in glomerular filtration rate, but may also be related to a neurogenic-mediated increase in renal tubular sodium reabsorption (26), or possibly related to an intrarenal shift of blood flow to the deeper juxtamedullary nephrons which

conserve sodium (2, 30). The trend for free water clearance to become less negative during continuous positive-pressure ventilation is related primarily to the decrease in osmolar clearance which occurred (31).

Thus, we have elucidated the reflex mechanism responsible for the antidiuresis associated with elevation of the expiratory pressure. Our data suggest that the application of 10 cm H_2O positive-end expiratory pressure produces hemodynamic changes on the arterial side of the circulation which decreases sinoaortic baroreceptor discharge activity (6). It appears that central nervous system integration of the afferent nerve signals from the baroreceptors results in an increased sympathetic discharge to the kidneys. The increased renal sympathetic nerve activity decreases renal blood flow and glomerular filtration rate which produces an antidiuresis and an antinatriuresis.

Summary. A study was conducted to describe the role of the renal nerves in mediating the reflex antidiuresis and antinatriuresis associated with the application of continuous positive pressure ventilation (CPPV). Experiments were performed on seven dogs 4–5 days after they had undergone surgical denervation of the left kidney. The nerves to the right kidney were left intact. Urine flow (V), sodium excretion ($U_{NA}V$), effective renal plasma flow (ERPF), and glomerular filtration rate (GFR) were determined for both the intact and denervated kidneys. In the right kidney with intact renal nerves, application of CPPV caused statistically significant ($P < 0.05$) decreases from control levels in V, $U_{NA}V$, ERPF, and GFR. In the left denervated kidneys, however, CPPV did not produce any statistically significant changes in V, $U_{NA}V$, ERPF, or GFR. These data suggest that the renal nerves participate in mediating the antidiuresis and antinatriuresis associated with elevation of the expiratory pressure.

1. Drury, D. J., Henry, J. P., and Goodman, J., J. Clin. Invest. **269,** 945 (1947).
2. Hall, S. V., Johnson, E. E., and Hedly-White, J., Anesthesiology **41,** 452 (1954).
3. Ueda, H., Neclerio, M., Leather, R. P., and Powers, S. R., Surg. Forum. **23,** 209 (1972).
4. Gauer, O. H., Henry, J. P., and Sieker, H. O., Prog. CV Dis. **4,** 1 (1961).
5. Murdaugh, H. V., Sieker, H. O., and Manfredi, F., J. Clin. Invest. **38,** 834 (1959).
6. Fewell, J. E., and Bond, G. C., The Physiologist **20,** 29 (1977).
7. Baratz, R. A., Philbin, D. M., and Patterson, R. W., Anesthesiology **34,** 510 (1971).
8. Tarak, T. K., and Chaudhury, R. R., Clin. Sci. **20,** 407 (1965).
9. Kezdi, P., and Geller, E., Amer. J. Physiol. **214,** 427 (1968).
10. Iriuchijima, J., and Wilson, M. F., Proc. Soc. Exp. Biol. Med. **131,** 189 (1969).
11. Katz, M. A., and Shear, L., Nephron **14,** 246 (1975).
12. Feigl, E. O., and D'Alecy, L. G., J. Appl. Physiol. **32,** 152 (1972).
13. Abe, Y., Dixon, F., and McNay, J. L., Amer. J. Physiol. **219,** 986 (1970).
14. Shipley, R. E., and Study, R. S., Amer. J. Physiol. **167,** 676 (1951).
15. Heyrovsky, A., Clin. Chim. Acta **1,** 470 (1956).
16. Waugh, W. H., and Beall, P. T., Kidney Int. **5,** 429 (1974).
17. Winer, B. J., "Statistical Principles in Experimental Design," 2nd ed. pp. 261. McGraw-Hill, New York (1971).
18. Steele, R. G. D., and Torrie, J. H., "Principles and Procedures of Statistics," pp. 481. McGraw-Hill, New York (1960).
19. Zar, J. H., "Biostatistical Analysis," pp. 121. Prentice-Hall, Englewood Cliffs, N. J. (1974).
20. Gammanpila, S., Bevan, D. R., and Bhudu, R., Brit. J. Anaesth. **49,** 199 (1977).
21. Knoefel, P. K., Handley, C. A., and Huggins, R. A., Proc. Soc. Exp. Biol. Med. **82,** 430 (1953).
22. Handley, C., and Moyer, J., J. Pharmacol. Exp. Ther. **112,** 1 (1954).
23. Katz, M. A., and Shear, L., Nephron **14,** 390 (1975).
24. Mercer, P., Canad. J. Physiol. Pharmacol. **49,** 420 (1971).
25. Berne, R. M., Amer. J. Physiol. **171,** 148 (1952).
26. Dibona, G. F., Amer. J. Physiol. **233,** F73 (1977).
27. Tucker, H. J., and Murray, J. F., J. Appl. Physiol. **34,** 573 (1973).
28. Kiil, F., Aukland, K., and Refsun, H. E., Amer. J. Physiol. **201,** 511 (1961).
29. Knox, F. G., Fleming, J. S., and Rennie, D. W., Amer. J. Physiol. **210,** 751 (1966).
30. Thurau, K., Proc. R. Soc. Med. **62,** 8 (1969).
31. Orloff, J., Wagner, H. W., and Davidson, D. G., J. Clin. Invest. **37,** 458 (1958).

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