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THE EFFECT OF DISTENDING THE ATRIAL APPENDAGES ON URINE FLOW IN THE DOG

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SUMMARY

1. Distension of the atrial appendages resulted in a diuresis, an increase in the rate of Na^+ excretion and an increase in heart rate.

2. Both the urinary and heart rate responses to distension of the appendages were either abolished or much reduced by crushing the bases of the appendages.

3. The diuresis in response to distension of the atrial appendages is similar to that previously described in response to distension of the pulmonary vein–atrial junctions by Ledsome & Linden (1968).

4. It is concluded that stimulation of nerve endings within the atrial appendages results in a reflex increase in urine flow and heart rate.

INTRODUCTION

Two types of afferent nerve endings have been demonstrated in the atrial endocardium; complex unencapsulated endings, the atrial receptors, and a system of fine nerve endings which has been called an end-net (Nonidez, 1941; Holmes, 1957; Miller & Kasahara, 1964). The atrial receptors are most numerous at the junctions of the veins and atria and have not been found in the atrial appendages whereas the end-net has been found throughout the atrial endocardium (Holmes, 1957; Miller & Kasahara, 1964).

Stimulation of atrial receptors by distension of small balloons placed at the pulmonary vein-left atrial junctions results in both a reflex increase in heart rate and a reflex increase in urine flow (Ledsome & Linden, 1964; Ledsome & Linden, 1968). Also stimulation of nervous structures within the atrial appendages by distension of balloons in the appendages results in a reflex increase in heart rate (Kappagoda, Linden & Saunders, 1972*a*). The present investigation was carried out to determine the effect of distending the atrial appendages on urine flow. A preliminary report of this investigation has been given (Kappagoda, Linden & Snow, 1972*c*).

METHODS

Dogs weighing 16·5–22·5 kg were given a s.c. injection of morphine sulphate (0·5 mg/kg) and 1 hr later under local anaesthesia (Decicaine 2%) a catheter was introduced through a saphenous vein into the inferior vena cava. The dogs were anaesthetized with an I.V. infusion of a solution of α-chloralose (dose 0·12 g/kg; Establissments Kuhlmann, Paris). Subsequently during the experimental procedures a steady state of light anaesthesia was maintained by further I.V. infusions of chloralose (approximately 0·01 g/kg every 10 min).

Two anaesthetic solutions were used; solution *a* consisting of 0·6 g NaCl and 1 g α-chloralose dissolved in 100 ml. distilled water and solution *b* consisting of 0·72 g NaCl, 0·21 g $NaHCO_3$ and 1 g α-chloralose dissolved in 100 ml. Dextraven 150 (Dextran 150 injection in 5% Dextrose, Fisons Pharmaceuticals Ltd, Loughborough). The initial dose of anaesthetic consisted of 75% of solution *a* and 25% of solution *b*; subsequently anaesthesia was maintained using solution *a*. Following the initial dose of anaesthetic 500,000 i.u. benzylpenicillin (Crystapen, Glaxo Laboratories Ltd, Greenford) were given I.V.

As soon as possible after the induction of anaesthesia the trachea was cannulated and artificial respiration started with 40% O_2 using a modified Starling 'Ideal' pump (Ledsome, Linden & Norman, 1967). When the chest was opened a resistance to expiration was provided by placing the outlet from the pump under 3 cm H_2O. Each ureter was catheterized through a flank incision and the urine volume measured every 10 min. Urinary sodium concentration was measured using a sodium electrode (BH 104 glass electrode; Electronic Instruments Ltd, Richmond, Surrey). The electrode was calibrated with gravimetrically prepared solutions of sodium chloride over the range 10–200 mM. The results obtained by this method for urinary sodium are slightly lower than those obtained by the flame photometer (Moore & Wilson, 1963). However, changes in urinary sodium concentration are reliably indicated by the electrode. The chest was opened in the mid line and retracted laterally by four hooks placed at each end of the sternum. The pericardium was opened and a small latex balloon about 5 mm long coated with silicone (Repelcote; Hopkin & Williams, Chadwell Heath, Essex) attached to a nylon catheter (1 mm i.d.) was inserted into each atrial appendage. Care was taken to cause as little surgical trauma as possible to the appendage during the above procedure; each appendage was opened by a small incision at its tip and the balloon positioned and tied within the tip of the appendage. Each appendage could then be distended by the injection of 4–5 ml. of a solution of NaCl (0·9 g/100 ml.) at a temperature of 38° C.

Pressures in the cardiovascular system were recorded through short (about 10 cm) nylon catheters (Portex surgical quality, size No. 4; Portland Plastics Ltd, Hythe, Kent) inserted into the right femoral artery, the right atrium through the right external jugular vein and the left atrium through the left upper pulmonary vein. Pressure in the trachea was also recorded. The methods of recording pressures, heart rate, e.c.g. and the end tidal P_{CO_2} have been described previously (Kappagoda, Linden & Snow, 1972*b*). The œsophageal temperature was measured using a telethermometer probe (Yellow Springs Instrument Co. Inc., Ohio, U.S.A.) and maintained at 37·5° C (± 1°) by adjusting heating lamps above and below the animal.

Samples of femoral arterial blood were withdrawn anaerobically at intervals throughout the experiment and the pH, P_{CO_2} and P_{O_2} measured as described by Ledsome, Linden, Norman & Snow (1967). The P_{CO_2} and pH were maintained within normal limits by adjusting the ventilation and by the continuous infusion of a solution of $NaHCO_3$ at a rate of 0·1 m-equiv $kg^{-1}.min^{-1}$.

Experimental protocol

Throughout the whole experiment urine was collected from each kidney and the volume during each period of 10 min was recorded. Half way through each collection period a constant volume of a mixture of two solutions was given I.V. The solutions were the anaesthetic solution *a* and a solution of NaCl (0·6 g/100 ml.). By altering the proportion of the two solutions the depth of anaesthesia could be varied as required and the total volume of fluid given every 10 min kept constant. The maximum maintenance dose of anaesthetic required was estimated during the initial part of each experiment. This dose then determined the total volume of fluid given every 10 min.

After the surgical procedures had been completed and when a steady state had been reached for at least 40 min with respect to anaesthetic dose, heart rate, arterial blood pressure and urine flow the atrial appendages were distended for a period of 30 min by injecting 4–5 ml. warm saline solution into each balloon. After the release of distension records were obtained and urine collected for a further 40 min.

Since it has been shown previously that the changes in urine flow in response to stimulation of nerve endings in the heart is delayed in onset (Ledsome & Linden, 1968) the mean control urine flow and urinary excretion of sodium were calculated as the mean of the three 10 min collections before distension and the final three 10 min collections of the 40 min period after distension. The mean urine flow and urinary excretion of sodium in response to distension of the appendages was calculated as the mean of the final two 10 min collections of the 30 min period of distension and the first 10 min period after the release of distension. The differences between the two calculated values for both urine flow and sodium excretion rate were taken as the response to distension of the appendages. This method of calculation of the responses is the same as that previously used (Henry & Pearce, 1956).

Changes in heart rate brought about by distending the vein-atrial junctions on either side of the heart are much more rapid in onset (Ledsome & Linden, 1964; Kappagoda *et al.* 1972*b*). Therefore changes in heart rate were calculated as the difference between the mean heart rate during the 30 min period of distension and the mean of the 30 min control periods immediately before and after the period of distension.

In order to destroy nerve fibres passing from the atrial appendages the base of each appendage was crushed with clamps for a period of at least 5 min. Such a technique has been demonstrated to abolish the reflex response of an increase in heart rate observed during distension of the atrial appendages (Kappagoda *et al.* 1972*a*). In each dog at the end of the experimental procedure the left atrium was distended by injecting a large volume of saline (15–20 ml.) into the balloon in the left atrial appendage for a period of 30 min and the changes in urine flow and composition, heart rate and blood pressures recorded and calculated as described above.

RESULTS

When recording began about 2 hr after the initial dose of anaesthetic the pH, P_{CO_2} and P_{O_2} of the arterial blood were 7·388 (mean; range 7·31–7·41); 37·4 mm Hg (mean; range 34–44); 223 mm Hg (mean; range 152–261) respectively. The heart rate was 101 beats/min (mean; range 68–136) and the mean arterial pressure was 140 mm Hg (mean; range 131–154).

The effect of distending the atrial appendages on urine flow

When a steady state with respect to urine flow, heart rate, arterial blood pressure and anaesthetic dose had been achieved, which occurred 2–5 hr after the completion of the surgical procedures, distension of the appendages a total of thirteen times in six dogs resulted in an increase in urine flow on each occasion.

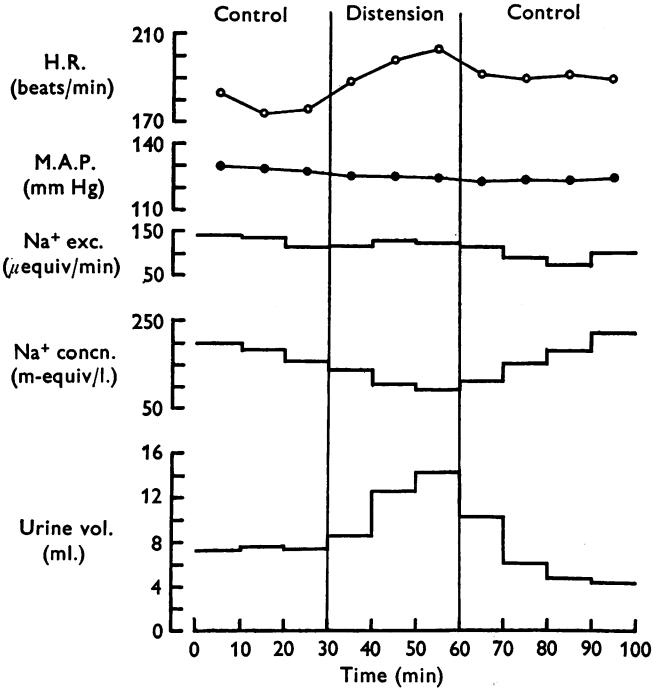

Fig. 1. The effect of distension of the atrial appendages in one dog (dog no. 109). From above down: heart rate (H.R.); mean arterial pressure (M.A.P.); urinary sodium excretion rate (Na^+ exc.); urinary sodium concentration (Na^+ concn.). The urine was collected over 10 min periods and the values for heart rate and mean arterial pressure refer to average values during the corresponding collection periods.

The response to distension of both atrial appendages in one dog is shown in Fig. 1. The mean urine flow in the control period before distension was 0·74 ml./min and increased to a mean flow of 1·23 ml./min. These changes in urine flow were accompanied by a change in the urinary sodium concentration which decreased during the period of distension as urine flow increased. During the period of distension there was a mean increase in heart rate of 14 beats/min. There were no significant changes in either the mean arterial pressure or the left and right atrial pressures (not shown in Fig. 1).

The changes in urine flow brought about by distending the appendages in all six dogs are shown in Fig. 2. The mean increase in urine flow calculated as a percentage of the control value was 23·2% (S.E. of mean ± 6·6) and is significantly different from zero ($0{\cdot}005 > P > 0{\cdot}001$; *t* test). The maximum increase was 100% and the minimum 7%. The changes in urinary sodium concentration and excretion rate during distension are shown in Fig. 3. In each dog the urinary sodium concentration was not always constant during the initial control period; on five occasions the

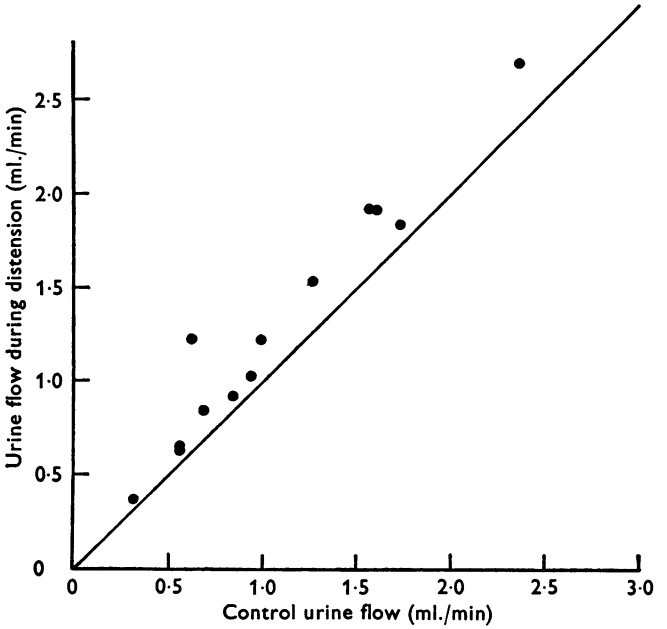

Fig. 2. The effect of distension of atrial appendages on urine flow. The urine flow (ml./min) during the period of distension plotted against the urine flow during the control periods (ml./min). Each point represents a single test and the continuous line is the line of no change.

sodium concentration was increasing, on five occasions steady and on three occasions falling. There is a significant fall in sodium concentration of 8·0% (mean; S.E. of mean ± 3·4; $0{\cdot}05 > P > 0{\cdot}025$; *t* test), and a significant increase in urinary sodium excretion rate of 11% (mean; S.E. of mean ± 2·4; $P < 0{\cdot}001$; *t* test). There were no significant changes in pressure in the right and left atrium during distension. The control pressure in the right atrium was 7·5 cm H_2O (mean; S.D. ± 1·7) and during distension was 7·4 cm H_2O (mean; S.D. ± 1·9). The control pressure in the left atrium was 8·0 cm H_2O (mean; S.D. ± 5·1) and during distension was 6·1 cm H_2O (mean; S.D. ± 4·8). The control arterial pressure was 135·4 mm Hg (mean; S.D. ± 10·3) and during distension was 134·9 mm Hg (mean;

S.D. ± 9·5) showing no significant change. There was however an increase in heart rate from 115·7 beats/min (mean; S.D. ± 31·9) during the control period to 129·7 beats/min (mean; S.D. ± 40·7) during the period of distension; an increase of 14·0 beats/min which is significant ($0{\cdot}01 > P > 0{\cdot}005$; t test; paired observations).

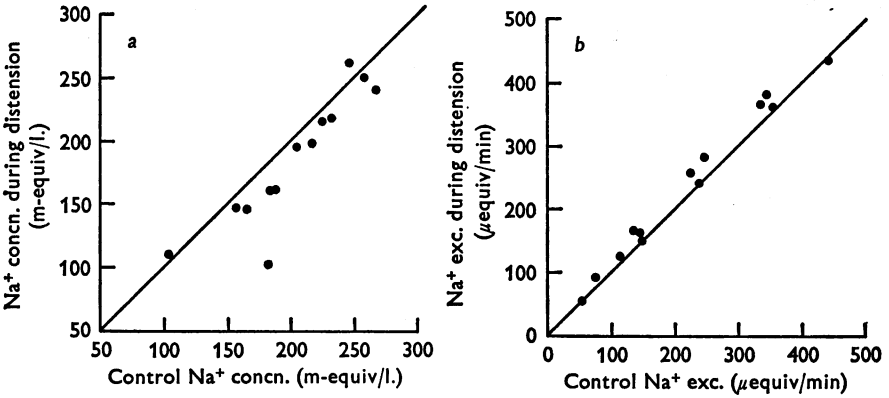

Fig. 3. *a*. The effect of distension of atrial appendages on urinary sodium concentration (m-equiv/l.) during the period of distension is plotted against the urinary sodium concentration (m-equiv/l.) during the control periods. *b*. The effect of distension of the atrial appendages on the urinary excretion rate of sodium. The urinary sodium excretion (μequiv/min) during distension is plotted against the urinary sodium excretion (μequiv/min) during the control periods. Each point represents a single test and the continuous line is the line of no change.

Effect of crushing the bases of the atrial appendages on the response to distension

In three dogs, after an initial response to distending the appendages had been obtained, the base of each appendage was crushed as described. Results obtained in one dog are shown in Fig. 4. Before the base of each appendage was crushed the response to distension was an increase in urine flow of 100% accompanied by a fall in urinary sodium concentration. After crushing the base of both appendages the response was reduced; the increase in urine flow was 22% accompanied by a small fall in urinary sodium concentration. The response of an increase in the heart rate to distension was also reduced by crushing the base of both appendages. In the other dogs both the increase in urine flow and the increase in heart rate were abolished. The results obtained are shown in Table 1.

In five dogs, including the three in which the bases of the appendages were crushed, the ability to produce an increase in urine flow in response to distension of the left atrium was tested by injecting a volume of between 15 and 20 ml. into the balloon in the appendage. The increase in urine flow was 89% (mean; range 13–185). There was a decrease in urinary sodium

concentration of 33% (mean; 16–43) and an increase in excretion rate of 23% (mean; 0–34). These changes in the amount and composition of the urine were accompanied by an increase in left atrial pressure from 12·0 cm H_2O (mean; range 7–15) to 31·0 cm H_2O (mean; range 20–42); and a fall in mean systemic arterial pressure from 135·5 mm Hg (mean; range 112–189) to 110·0 mm Hg (mean; range 90–124). The above changes are similar to those in response to distension of the left atrium previously reported by Henry & Pearce (1956), Ledsome, Linden & O'Connor (1961) and Ledsome & Linden (1968).

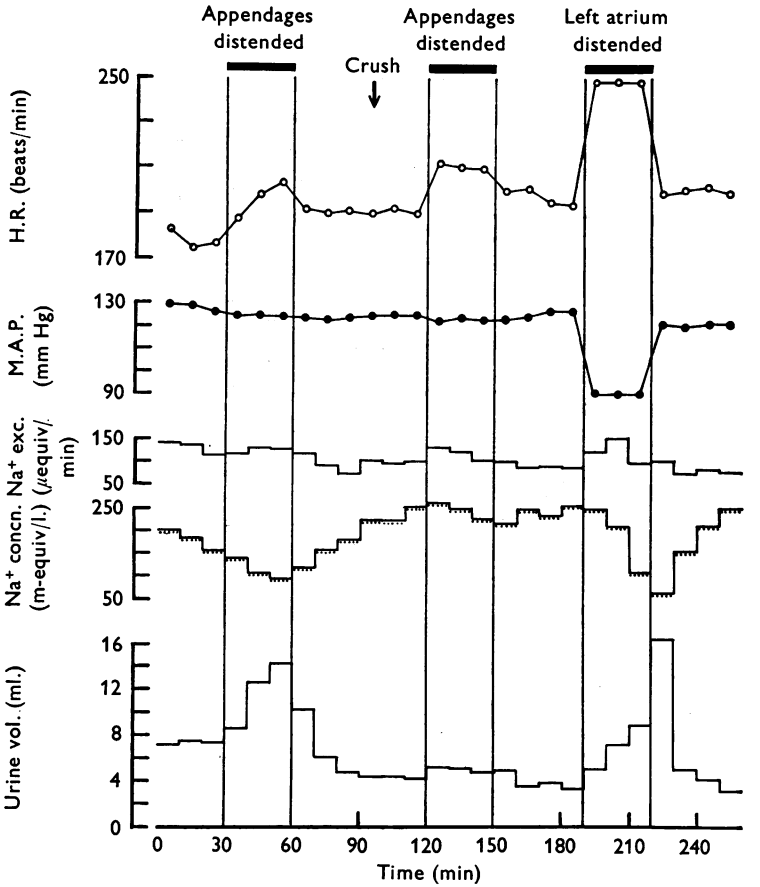

Fig. 4. The effect of crushing the bases of both atrial appendages on the response to distension of the atrial appendages; and the effect of distension of the left atrium. From above down heart rate (H.R.); mean arterial pressure (M.A.P.); urinary sodium excretion (Na^+ exc.); urinary sodium concentration (Na^+ concn.); urine volume. The appendages were distended before and after crushing the bases of the appendages. The left atrium was distended after the completion of the above.

TABLE 1. Effect of crushing the bases of the atrial appendages on the response to distensions of the atrial appendages; and the effect of distension of the left atrium

Dog no.	Urine flow (ml./min)			Urinary [Na^+] (m-equiv/l.)			Na^+ excretion rate (μequiv/min)			Heart rate (beats/min)		
	Control	Test	Diff.	Control	Test	Diff.	Control	Test	Diff.	Control	Test	Diff.
108 *A*	0·69	0·84	+0·15	104	110	+6	72	92	+20	132	150	+18
B	0·50	0·50	0	101	101	0	51	51	0	145	145	0
C	0·47	0·53	+0·06	90	76	−14	42	40	−2	155	216	+61
109 *A*	0·62	1·23	+0·61	183	104	−79	113	128	+15	180	192	+12
B	0·40	0·49	+0·09	237	229	−8	95	112	+17	194	204	+10
C	0·38	1·08	+0·70	224	127	−97	85	137	+52	198	248	+50
112 *A*	0·81	0·92	+0·11	186	165	−21	151	152	+1	70	73	+3
B	0·72	0·70	−0·02	288	303	+15	207	212	+5	77	76	−1
C	0·84	1·59	+0·75	281	201	−80	236	320	+84	91	169	+78

A. Distension of both appendages before crushing.
B. Distension of both appendages after crushing.
C. Distension of the entire left atrium after crushing.

DISCUSSION

Complex unencapsulated nerve endings (atrial receptors) have been demonstrated on the endocardial surface at the entrances of the veins into both atria (e.g. Nonidez, 1937). Stimulation of these atrial receptors results in a reflex increase in heart rate (Ledsome & Linden, 1964; Kappagoda *et al.* 1972*b*). The afferent path of this reflex is in the vagi and the efferent solely in the sympathetic nerves to the heart. Ledsome & Linden (1968) have also shown that stimulation of the left atrial receptors results in a diuresis which is abolished by sectioning the vagi in the chest. On the basis of these findings Ledsome & Linden (1968) concluded that the atrial receptors played a major role in the diuresis which followed distension of balloons in the cavity of the left atrium (Henry & Pearce, 1956; Ledsome *et al.* 1961).

The time course of the response of an increase in urine flow obtained in the present investigation is similar to that observed by Henry & Pearce (1956), Ledsome *et al.* (1961) and Ledsome & Linden (1968), when the whole of the left atrium was distended and also similar to the response observed by Ledsome & Linden (1968) when only the pulmonary vein-left atrial junctions were distended. The magnitude of the response though smaller than that observed when the whole of the left atrium is distended, is similar to that observed in response to distension of the pulmonary vein-left atrial junctions. It is therefore suggested that the mechanism by which all three methods produce a diuresis is the same, i.e. all three methods of distending the atria stimulate the same type of receptors.

However, it is important to remember that at least three investigations have stated emphatically that there are no unencapsulated nerve endings in the endocardium of the atrial appendages of dog and man, only the end-net has been demonstrated (Holmes, 1957; Miller & Kasahara, 1964; Johnston, 1968). In spite of the reputed absence of the complex unencapsulated endings in the endocardium of the atrial appendages of the dog (Nonidez, 1941; Holmes, 1957; Miller & Kasahara, 1964), Kappagoda *et al.* (1972*a*) showed that distension of the atrial appendages without causing a change in atrial pressures resulted in a reflex increase in heart rate which was qualitatively similar to that caused by stimulation of the atrial receptors at the vein-atrial junctions. Kappagoda *et al.* (1972*a*) have also shown that the reflex increase in heart rate caused by the distension of the atrial appendages was abolished by crushing the bases of the appendages.

In the present study the same procedure either abolished or diminished both the increase in heart rate and the diuresis. It could be concluded therefore that the diuresis is a reflex response and that the receptors responsible for both these responses are located in the atrial appendages. There are several types of receptor end-organs which could be involved

either individually or collectively in these reflexes. First, it is possible that complex unencapsulated endings are present in the endocardium of the appendages but have remained undetected. Secondly, the so-called end-net could be a receptor end-organ. Again it is at least possible that both the complex unencapsulated endings and the end-net might be involved in these responses. Finally, receptors located at sites other than the endocardium could mediate these responses, e.g. epicardial receptors (Sleight & Widdicombe, 1965).

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