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REFLEX CARDIOVASCULAR AND RENAL RESPONSES FROM THE PULMONARY ARTERIES OF THE ANESTHETIZED DOG

BY

WAI-ON KAN

B.Sc. (DISTINCTION), WASHINGTON STATE UNIVERSITY, 1970

M.Sc., UNIVERSITY OF SOUTHERN CALIFORNIA, 1973

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Department of Physiology

The University of British Columbia
2075 Wesbrook Place
Vancouver, Canada
V6T 1W5

Date April 6, 1977.

ABSTRACT

A preparation is described utilizing a constant flow, right-heart bypass for perfusion of the isolated main pulmonary arteries at controlled pressures. It is demonstrated that stepwise increments of pressure in the pulmonary arteries are accompanied by increases in systemic vascular resistance and in hind-limb vascular resistance. These changes were demonstrated over the whole range of 5-120 cm H_2O pressure in the pulmonary arteries. In contrast there were no significant changes in renal vascular resistance or heart rate. It is also shown that changing the temperature of the perfusate in the pulmonary artery from 37°C to 30°C is associated with a decrease in systemic vascular resistance. The effects of raising the pulmonary arterial pressure and of cooling the pulmonary artery were abolished by cervical vagotomy. It is suggested that there is a tonic reflex vasoconstrictor tone generated by activity of receptors lying in or close to the walls of the pulmonary artery. It is further suggested that the differential effects on systemic vascular resistance and renal resistance may distribute cardiac output preferentially to the kidney providing one mechanism by which changes in blood volume may lead to appropriate changes in renal solute excretion. The later hypothesis was put to test by collecting urine from the intact kidneys of animals with isolated pulmonary pouch preparation. A step increase in the pouch pressure evoked a corresponding rise in the urinary volume, osmolar

clearance, sodium excretion rate, but not the free water clearance and the potassium excretion rate. The response may be caused by renal hemodynamic changes as a result of the reflex increase in systemic arterial pressure. The rise in sodium excretion rate continued even after the release of the pressure in the pulmonary artery pouch thus the role of a natriuretic hormone in the reflex was suspected. A series of animals with one kidney intact and one kidney isolated and perfused with constant pressure was used in attempt to demonstrate the natriuretic action. These results confirmed the hemodynamic effect on the urinary function of the intact kidney. In the isolated kidney there was no statistically significant increase in sodium excretion rate, therefore the role of a natriuretic agent in the reflex response to distension of the pulmonary artery is still uncertain.

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INTRODUCTION

J. P. Peters (1935) first proposed a principle of circulatory blood volume control. He reasoned that the fullness of the blood stream may provoke diuretic response on the part of the kidney. Many groups of investigators have verified this concept experimentally by isosmotic expansion of blood volume, among them Borst (1948), Zuidema, Clarke, Reeves, Gauer and Henry (1956) recorded an increase in urine flow after the expansion. Analysing the distensibility properties throughout the circulation Gauer and Henry (1963) adapted the terms low- and high-pressure systems to distinguish the pulmonary circulation, right heart and capacitance vessels from the systemic arterial circulation. Because of the elastic resistance of the low-pressure circuit is 100 to 200 lower than the high-pressure circuit they reasoned that during a blood volume expansion the majority of the extra volume would go to the low-pressure circuit. From a physical point of view biological receptors registering vascular bed tension at any region throughout the low-pressure system could sense the "fullness of the blood stream", and function as "volume receptors".

Efforts to locate the receptors in the low-pressure system have resulted in further functional subdivision into intrathoracic and capacitance vessel compartments. Different types of maneuvers to alter the intrathoracic blood volume such as high positive pressure breathing (Drury,

Henry and Goodman, 1947; Murdaugh, Sieker and Manfredi, 1959), negative pressure breathing (Sieker, Gauer and Henry, 1954; Pabst and Gauer, 1959), water immersion (Arborelius, Balldin, Lilja and Lundgren, 1972; Lange, Lange, Echt and Gauer, 1974) and saline infusion (Strauss, Davis, Rosenbaum and Rossmeisl, 1951; Levinsky and Lalone, 1963) were employed to investigate the intrathoracic area. The general result of all these different experiments was that an expanded intrathoracic volume would bring on a diuresis and natriuresis, while a decreased intrathoracic volume is followed by oliguria. Since there are no proven reflex activities originating from the capacitance vessels it is widely accepted that the intrathoracic compartment is a reflexogenic area which regulates blood volume.

Electrophysiological recordings of the rhythmic activity in vagal afferents from the heart have shown a population of nerve endings on the low-pressure side of the circulation functioning as receptors (Paintal, 1953). In the case of some left atrial receptors the stimulus appears to be related to the degree of stretch of the atrial muscle fibers thus the intrathoracic blood volume. Distension of the left atrium by inflating a balloon has produced an unequivocal diuretic response (Ledsome, Linden and O'Connor, 1961; Shu'ayb, Moran and Zimmermann, 1965; Johnson, Moore and Segar, 1969). The diuresis although challenged by some investigators has been shown by good evidence to be mediated

through the control of ADH release (Ledsome and Mason, 1972; De Torrente, Robertson, McDonald and Schrier, 1975). The ADH mechanism explained the increase in free water clearance associated with an expanded intrathoracic blood volume but it can not account for the accompanying small natriuresis. Other sites in the intrathoracic area have also been examined for possible reflexogenic effects.

The main pulmonary artery and its bifurcations is richly supplied with sensory nervous endings. It is developed from the VI embryonic visceral arch (Koch, 1931) and structurally similar to the carotid sinus and aortic arch baroreceptor sites. Since the first histological study by Dogiel (1898) nervous endings in conspicuous rings, with branching coils and fibers with bushy endings have been reported by many investigators (Nonidez, 1937; Coleridge, Kidd and Sharp, 1961; Grigor'eva, 1962), they are thought to be baroreceptors. Both sympathetic and parasympathetic innervations have been traced to the pulmonary artery receptors (Coleridge, Kidd and Sharp, 1961; Edgeworth, 1892; Grigor'eva, 1962) and the nervous connections can either be fast conducting large diameter myelinated A fibers or small myelinated or unmyelinated C fibers (Coleridge, Coleridge, Dangel, Kidd, Luck and Sleight, 1973). The large myelinated vagal fibers discharge actively at pressures normally present in the pulmonary artery. Besides these nervous endings Nonidez (1937) described some glomus tissue in the pulmonary artery adventitia which might function as chemoreceptors.

Attempts to stimulate the pulmonary artery receptors with hydraulic pressure have produced inconsistent results (Churchill and Cope, 1929; Schweitzer, 1936; Parin, 1947). Part of the inconsistency may be caused by pressure changes in the lungs when the pulmonary arterial pressure was increased. Improved techniques developed by Osorio and Russek (1962), Coleridge and Kidd (1963) allowed the pulmonary arterial pressure to be increased locally without impairment to the cardiac output. They reported two types of responses. Weak distension of the artery produced either no change or a fall in systemic pressure, while a stronger distension always produced a rise.

The existence of a large population of receptors in the pulmonary artery has been proven beyond any doubt. Their afferent pathways and activities under different pulmonary arterial pressures have also been closely examined, but so far their physiological function is still uncertain. The only consistent result known is an increase in the systemic pressure when the pulmonary artery is subjected to pressure above 80 mm Hg.

The following experiments were undertaken in the hope that a further improved experimental technique, providing better control of the physiological variables involved, would elucidate the reflex cardiovascular effects of pulmonary artery pressure changes and their possible role in the regulation of blood volume.

METHODS

The experimental design allowed three major reflex response properties originating from the pulmonary artery to be investigated. The first group of experiments were designed to demonstrate the changes in the hind-limb, kidney and the general systemic vascular resistance as a result of changes in the pulmonary artery pressure. In the second group the combined effects of cooling and distending the pulmonary artery and the role of the vagus nerves were illustrated. The last group of experiments showed the regulation of urine output when the pulmonary artery was subjected to different static pressure, and examined the behaviour of intact and isolated kidneys in response to the same pulmonary artery distension.

A. Anesthesia and General Preparation

Mongrel dogs of 20 to 35 kg were subcutaneously injected with morphine sulphate (0.5 mg/kg). One hour later under local anesthesia (mepivacaine hydrochloride, 1%) a polyethylene catheter was inserted through a saphenous vein into the inferior vena cava and each dog was anesthetized by chloralose (British Drug Houses). The chloralose was prepared in a solution of 0.9% NaCl at 60°C to make a 1 gm chloralose/100 ml NaCl solution. This solution was filtered to remove undissolved particles prior to use. The chloralose was infused through the saphenous cannula (0.1 gm/kg) to induce anesthesia. Subsequently, during the experimental procedures a steady state of light anesthesia and fluid input was maintained by the constant infusion of additional chloralose solution into the external jugular vein delivered by a motor driven syringe pump (Harvard Apparatus Co. Inc., Dover, Mass.). The compositions and infusion rates of the additional chloralose solutions are listed under the individual groups of experiments.

Body temperature of the animal was maintained around 37°C using a heated table controlled by a thermistor probe inserted into the esophagus, activating a telethermometer control unit (Yellow Spring Instruments Co., Inc.). Systemic arterial pressure was recorded through a 8 cm length of teflon tubing (1 mm bore) from either the right femoral artery or the right brachial artery. Elec-

trocardiogram was recorded from leads placed in the right leg and chest region. The signal was amplified by a pre-amplifier (Grass Instruments, Quincy, Mass., P15) and displayed on an ultraviolet recorder. Heart rate was calculated by counting the electrocardiogram record over periods of thirty seconds.

B. Right-Heart Bypass And Pulmonary Artery Pouch Preparation

As soon as possible after the induction of anesthesia artifical respiration was started via a tracheal cannula with a mixture of 40% oxygen in air, supplied from a respiration pump (Harvard Model 614) at a rate of 14 strokes per minutes and a stroke volume of approximately 13 ml/kg body weight. End tidal CO_2% was continuously monitored at the tracheal cannula by a CO_2 analyser (Beckman, Medical gas analyzer, Model LB-1). When the chest was opened a resistance to expiration equivalent to 3 cm H_2O was provided by an exhalation valve (Ohio Chemical Co.). During the surgical procedures, the dogs received a slow infusion of 100 ml of Dextran (6% Dextran 75 in 0.9% NaCl Travenol Inc.) for each 13 kg body weight (approximately 10% of their estimated blood volume).

Thoracotomy was performed by splitting the sternum in the midline. Succinylcholine chloride (Squibb Laboratories, 0.5 mg/kg) was given to the animals before the thoracotomy to eliminate muscular twitching which inter-

fered with the surgery. The skin and muscle were divided using electrocautery (Birtcher model 755 Electro-surgical unit), an electric surgical saw (Styker Corp. model 8381-210) was used to split the sternum. Hemostasis was achieved by tying and cutting the internal mammary arteries and veins, cauterizing and bone waxing the cut surfaces of the bone.

The left pulmonary artery was dissected free close to its point of bifurcation outside the pericardium at the root of the left lungs. A double lumen stainless steel cannula with an inner tube of 0.5 mm bore for pressure recording and an outer tube of 5 mm bore for perfusion was inserted into the left pulmonary artery. Right heart bypass was established by inserting a 1 cm bore polyethylene tubing into the right atrial appendage and advancing it into the right ventricle. Blood drained through the tubing into a constant temperature reservoir which was maintained at 37°C. From the reservoir, blood passed through an electromagnetic flow meter (Biotronics Inc. BL 610) and was pumped into the left lungs by a roller pump; constant flow and thus constant cardiac output being maintained using a negative feedback control signal from the electromagnetic flow meter (Fig. 1). Before opening the pericardium, propranolol (0.3 mg/kg, in 20 ml of 0.9% saline) was injected intravenously to reduce the tachycardia and arrhythmias which accompanied handling of the heart during the surgical procedure. A priming dose of heparin (500 units/kg i.v., heparin sodium 100 units

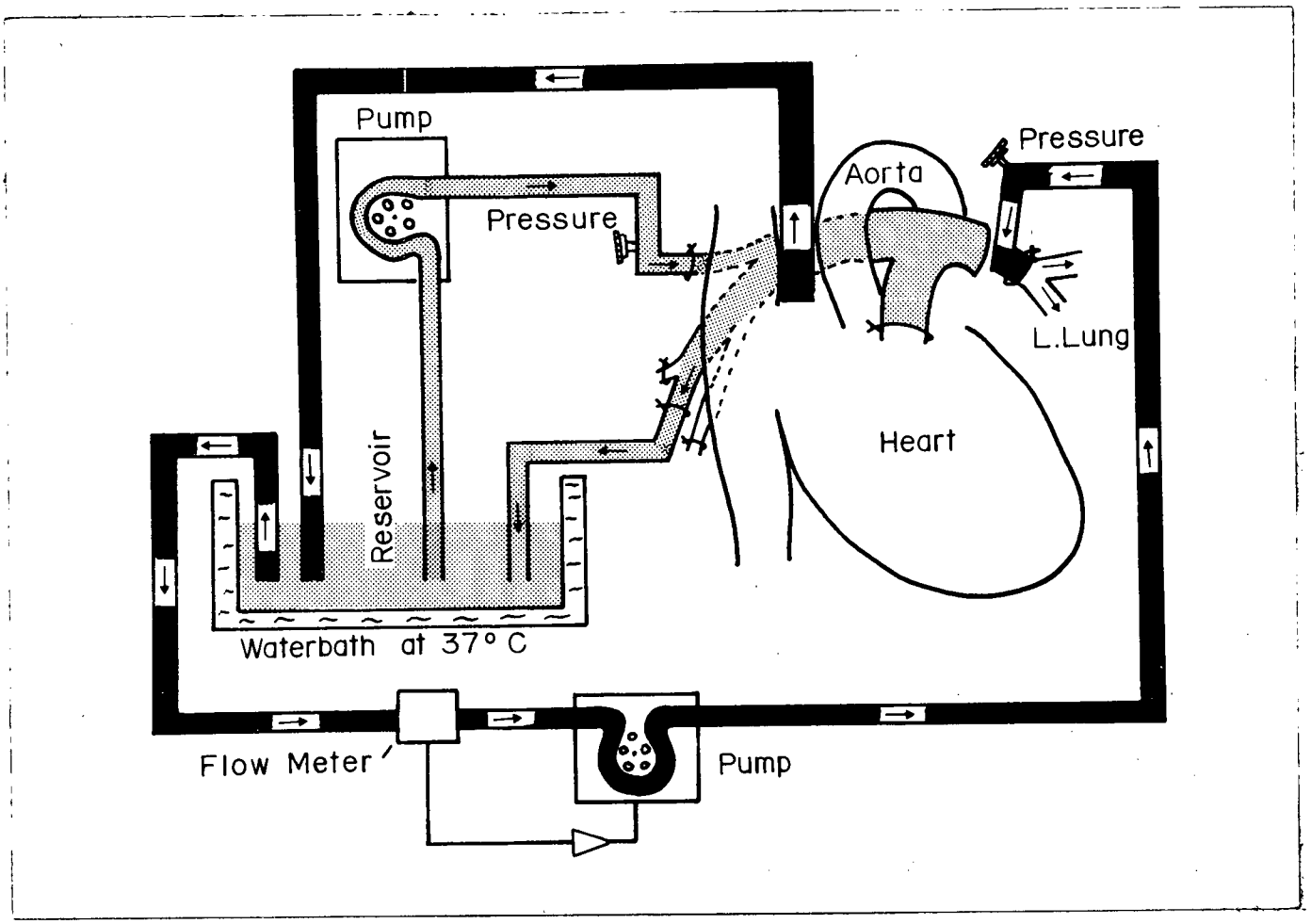

Fig. 1 Diagram of method of right heart bypass and perfusion of the isolated pulmonary arteries.

/mg, Nutritional Biochemicals Corp.) was injected prior to establishing the right heart bypass.

The root of the main pulmonary artery was carefully separated from the aorta and a snare placed around the pulmonary artery just above the pulmonary valve. Branches of the right pulmonary artery supplying the first and third lobes of the right lung were also dissected free; the root of the second and fourth lobes were tied tightly. The cannula in the first right pulmonary artery branch provided the inflow from the pump and was also a double lumen stainless steel cannula, an inner tube of 0.5 mm bore was used for pressure recording the larger outer tube of 3 mm bore for perfusion. The tip of the inflow cannula was placed close to the pulmonary artery bifurcation to insure thorough mixing of the perfusate in the artery, thus achieving an uniform temperature throughout the pouch. The outflow was through the third branch of the right pulmonary artery via a single lumen 3 mm bore stainless steel cannula. Thus the main pulmonary artery and its bifurcation became a closed pouch which was perfused with blood, or saline as specified in the different groups of experiments, by a roller pump which drew venous blood from the reservoir or warm saline from a separate reservoir. Pressure in the pulmonary arterial pouch could be varied by controlling the speed of the roller pump and by a screw clamp placed on the out-flow of the circuit.

The right heart bypass and pulmonary artery pouch were prepared in all the experiments described in this thesis.

The reservoir and tubing were primed with a mixture of 50% Dextran (6% Dextran 75 in 0.9% NaCl, Pharmacia or Travenol Inc.) and 50% Ringer Lactate solution (Travenol Inc.) the total priming fluid was either one, two or three liters depending on the experiment. During the experimental observations a maintenance dose of heparin (50 units/kg i.v.) was admistered every thirty minutes.

C. Hind-Limb And Kidney Perfusion

a) Hind-limbs

The hind-limbs were perfused in six dogs to investigate the effect of pulmonary arterial distension on the hind-limb vascular resistance. A right heart bypass was prepared as described before, then a flank incision was made on the left side exposing a segment of the descending aorta between the renal arteries and the common iliac arteries. Upon tying the inferior mesenteric and lumbar arteries in that section a 5 mm diameter stainless steel cannula was inserted rostrally below the renal arteries, the arterial blood then passed through a constant flow roller pump and an electromagnetic flow meter (Biotronics Inc. BL 610) and entered the animal through a double lumen cannula (5 mm bore outer tube, 0.5 mm bore inner tube for pressure recording) inserted caudally above the common iliac arteries (Fig. 2). The pump was adjusted such that the perfusion pressure of the hind-limb was approximately the same as the systemic pressure at the beginning of the experiment and the blood flow was then maintained constant throughout the experiment.

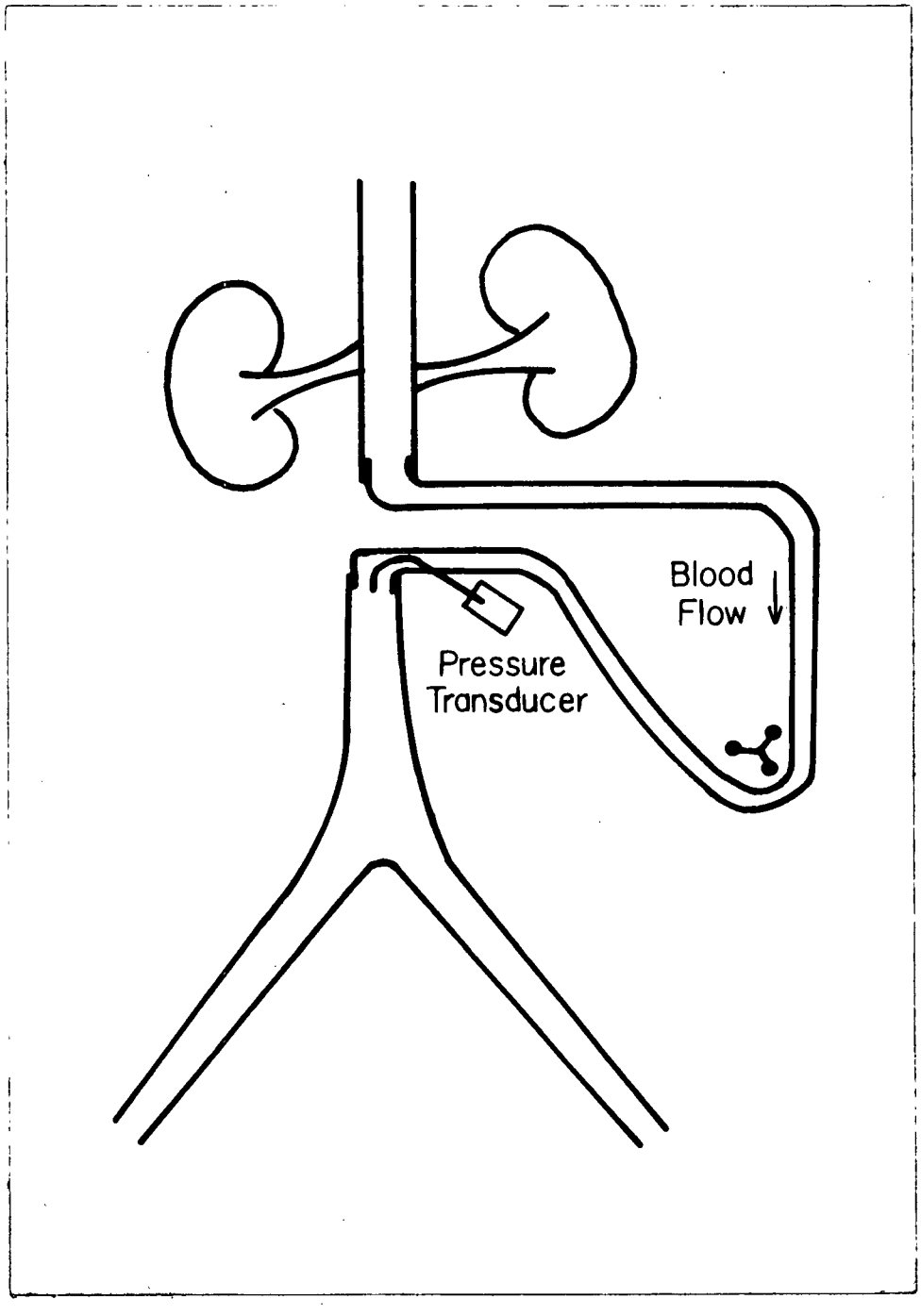

Fig. 2 Diagram of method of hind-limb perfusion.

For both the limb perfusion and kidney perfusion experiments the reservoir was primed with 1 liter of the Dextran and Ringer Lactate mixture. Continuous anesthesia was maintained by constant infusion of 0.5% chloralose solution (0.5 g chloralose in 100 ml of 0.9% sodium chloride solution) at a rate of approximately 1.0 ml/min. The pulmonary arterial pouch was perfused with blood at 37°C drawn from the reservoir except for two of the limb perfusion experiments in which the pulmonary arterial pouch was perfused with cool saline of different temperatures after completion of the normal experimental protocol to investigate its response to cooling. In another two of the limb perfusion experiments muscular jerks were observed, which are a feature of chloralose anesthesia, consequently succinyl choline was given as a continuous infusion at a rate of 0.5 mg/min together with the 0.5% chloralose.

b) Kidneys

In six dogs the changes in kidney resistance in response to pulmonary arterial pressure were examined. The descending aorta was dissected free in two places one about one inch above and the other two inches below the renal arteries. Testicular or ovarian and lumbar arteries in that section were tied and cut. A 5 mm diameter stainless steel cannula was inserted into the left subclavian artery directing arterial blood through a roller pump and an electromagnetic flow meter to the descending aorta.

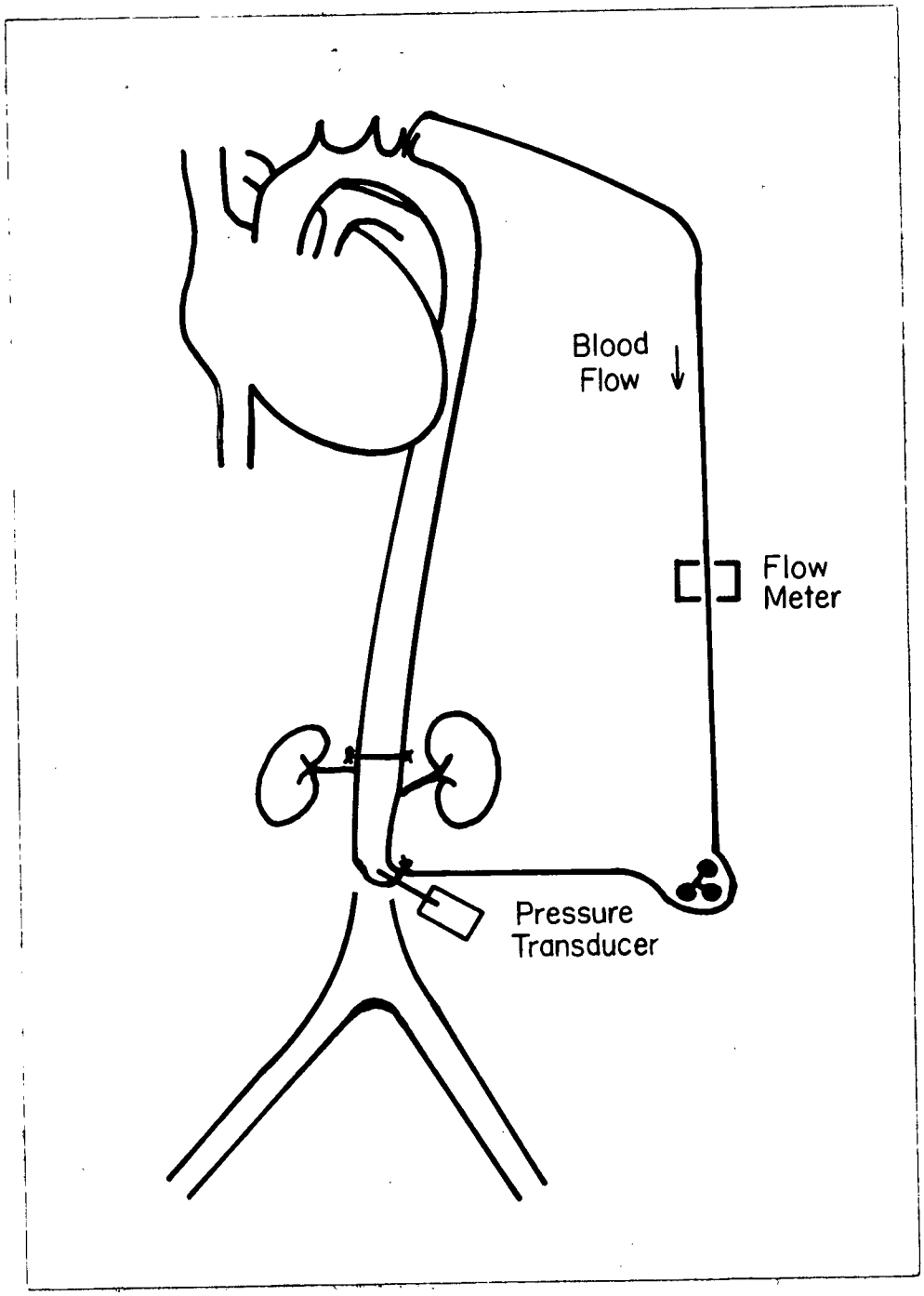

Fig. 3 Diagram of method of kidney perfusion.

rostrally below the renal arteries through a double lumen cannula (5 mm bore outer tube, 0.5 mm bore inner tube). The aorta was tied just above the renal arteries such that the kidneys were perfused solely by the pump (Fig. 3). Because the distance between the two renal arteries varies, in some dogs only one kidney was perfused. Renal perfusion pressure was measured through the 0.5 mm bore inner cannula. Pump speed was adjusted so that the renal arterial pressure was approximately the same as that of the systemic at the beginning of the experiment. Either constant flow (constant pump speed) or constant pressure (feedback control to the pump from the renal arterial pressure transducer) was possible. Both modes were used on separate occasions.

D. Cooling

Ten dogs were prepared with right heart bypasses as described before and the right femoral artery was cannulated for systemic arterial pressure recording. The constant temperature blood reservoir composition and volume were the same as the limb and kidney perfusion experiments. Continuous anesthesia was achieved by infusing 0.5% chloralose solution in 0.9% saline at about 1.0 ml/min. However, the pulmonary artery pouch was perfused with 0.9% saline at 30°C instead. Cool saline was used in this case in order to elucidate the combined effect of cooling and distending the pulmonary artery.

In eight of these ten dogs the vagus nerves were cut in the neck after completion of the experimental protocol and the experiment repeated. Before each experiment was started, the ability of the preparation to respond reflexly was checked by occluding both carotid arteries. By cutting the vagus nerves any part of the pulmonary arterial reflex which depends on them will be eliminated.

E. Urine Collection-Kidneys Intact

Twenty-nine dogs were prepared for urine collection from the intact kidneys. These dogs were divided into three groups in which the pulmonary artery pouches were subjected to different pressure manipulations. The blood reservoir was primed with 2 liters of 50% Ringer Lactate and 50% Dextran as described before except for the increased volume. In addition 40 ml of 1N $NaHCO_3$ was incorporated into the reservoir to bring the pH of the reservoir to approximately 7.4. Continuous anesthesia and volume replacement was maintained in this case by infusing a mixture of 500 ml of 0.5% chloralose in 0.9% saline and 80 ml of 1N $NaHCO_3$ delivered to the dog at approximately 2 ml/min.

Flank incisions were made on the left and right sides. The two ureters were dissected from the dorsal abdominal wall and cannulated approximately four inches below the kidneys. Cannulae used were about 30 cm long polyethylene tubings with 1.2 mm bore. The transversus

abdominis, internal and external oblique muscles and the skin were then sutured and urine produced by the left and right kidneys was collected separately for analysis. Urine samples were collected every ten minutes and a 5 ml blood sample was taken every twenty minutes and centrifuged immediately for a plasma sample.

The urine volume was measured with volumetric cylinders. Osmolarities of the plasma and the urine samples were measured with an osmometer (Osmette Precision Osmometer, Precision Systems) by the depression of freezing point method. To measure the sodium and potassium concentrations of the samples, they were first diluted (15 μl of sample to make up to 4 ml with standard Li solution) with an automatic diluter (Fisher Diluter, Model 240). The standard Li solution contained 15 mEq of Li per liter. Then a flame photometer (Instrumentation Laboratory Inc., Boston, Mass. Model 143) was used to analyse the diluted samples.

In ten of the dogs, the pulmonary arterial pouch pressure was kept constant at 15 cm H_2O throughout the entire experiment. Data obtained from these experiments serves as control values and forms the basis of comparison with the other two sets in this series. In the second set of nine dogs the pulmonary arterial pouch pressure was kept at 5 cm H_2O at the beginning of the experiment and raised to 40 cm H_2O for a period of 30 min then released

again to 5 cm H_2O. For the last ten dogs the pulmonary arterial pressure was 15 cm H_2O to start with then increased to 80 cm H_2O for 30 min before it was lowered back to 15 cm H_2O. Blood from the constant temperature reservoir was used to perfuse the pulmonary artery pouch. From the three sets of experiments we would be able to see if there was any graded response in the urine output and urine composition in response to stimuli of different magnitude from the pulmonary artery pouch.

F. Urine Collection-Intact And Isolated Kidneys

Urine composition and volume can be varied by many different factors. Among them renal arterial pressure, sympathetic nervous activity to the kidneys and blood borne agents exert major influences. An intact kidney will be subjected to the control of all the mentioned factors. However, for an isolated kidney perfused at constant renal arterial pressure the arterial pressure and sympathetic nervous activity factors will be eliminated, changes in the urine output and composition will be dicated by blood borne agents.

In six dogs the right kidneys were kept intact, and the ureters were cannulated as before. Flank incision was made high on the left side about half an inch below the last rib cutting transversely across the external and internal oblique muscles exposing the left kidney. The ure-

ter, renal artery and vein were dissected free and all the nerves and connective tissues surrounding the kidney were tied and cut. A 5 mm diameter stainless steel cannula was inserted into the left subclavian artery to draw arterial blood and then pumped into the left kidney through a 2 mm diameter "V" shaped cannula. Perfusion pressure was measured from a side arm of the cannula. The renal vein was cannulated with a 3 mm bore cannula and venous blood was drained into the reservoir. Urine output was collected through a 1.2 mm diameter polyethylene tubule cannulated into the ureter. After cannulation was completed the only connections of the kidney with the animal were through the perfusion cannulae. The kidney was placed back into the cavity created by the surgery and covered with swabs soaked with normal saline. In this position the kidney was warmed by the perfused blood and the surrounding tissue. Blood flow to the kidney was measured by an electromagnetic flow meter (Biotronic Inc. BL 610) and the perfusion pressure was kept constant by a feed-back signal from the pressure transducer which controls the roller pump.

The blood reservoir was primed with three liters of Dextran and Ringer Lactate mixture plus 60 ml of 1N $NaHCO_3$ to bring the pH up to about 7.4. For continuous anesthesia and fluid replacement and to maintain acid-base balance a mixture of 500 ml of 0.5% chloralose in 0.9% saline, 180 ml of 0.44N $NaHCO_3$ was infused at approximately 2 ml/min.

The pulmonary artery pouch was perfused with blood at body temperature. Its pressure was kept at 10 cm H_2O at the beginning of the experimental period then raised to 100 cm H_2O for 30 min. and lowered back to 10 cm H_2O afterwards. Urine and plasma samples were collected and analysed as before.

TABLE 1

Experiments Performed

Dog #	Weight kg	Type of expt.	Techniques
1	38	(Hind-limb	CLP
2	26	(perfusion	"
3	25		"
4	26		"
5	29		" , Pouch cooling
6	34		" , " "
7	28	(Kidney	CRP, CRF
8	30	(perfusion	CRF, CRF
9	29		CRP
10	28		CRF
11	26		CRP, Pouch cooling
12	27		CRP, Pouch cooling
13	30	(Pulmonary artery	Vagi intact, Vagi cut
14	29	(pouch perfusion	" " , " "
15	35	(with 30°C saline	" "
16	35		" "
17	32		" " , Vagi cut
18	30		" " , " "
19	22		" " , " "
20	26		" " , " "
21	23		" " , " "
22	22		" " , " "

CLP = Constant limb blood pressure
CRP = Constant renal blood pressure
CRF = Constant renal blood flow

TABLE 1 (continued)

Dog #	Weight kg	Type of expt.	Techniques
23	24	(Urine collection	Both kidneys intact
24	25	(control	
25	28		
26	24		
27	28		
28	30		
29	28		
30	28		
31	24		
32	27		
33	25	(Urine collection	Both kidneys intact
34	25	(pulmonary artery	
35	24	(pouch distended	
36	30	(to 40 cm H_2O	
37	25		
38	28		
39	35		
40	25		
41	27		
42	25	(Urine collection	Both kidneys intact
43	22	(pulmonary artery	
44	25	(pouch distended	
45	30	(to 80 cm H_2O	
46	28		
47	27		
48	23		
49	26		
50	25		
51	33		

TABLE 1 (continued)

Dog #	Weight kg	Type of expt.	Techniques
52	25	(Urine collection	(Right kidney
53	23	(pulmonary artery	(intact,
54	22	(pouch distended	(left kidney
55	21	(to 100 cm H_2O	(isolated
56	25		
57	21		

EXPERIMENTAL PROTOCOL

After completion of the surgical procedures, the animal was allowed to stablize for a period of time (15 min. minimum for the limb, kidney perfusion and pulmonary artery cooling experiments; 30 min. minimum for the urine collection experiments) and during this time a sample of the arterial blood was taken and pH, PCO_2 and PO_2 were measured using appropriate electrodes (Instrumentation Labortories, Model 113-51). If necessary, adjustments were made to the respiratory pump or small infusions (10-30 mEq of sodium bicarbonate solution, 1N) were given to maintain an arterial PCO_2 between 35 mm and 40 mm of mercury and pH within the range of 7.35 to 7.42; no adjustments were made during the subsequent control or experimental periods.

To each pressure recording cannula was attached a Statham strain gauge (Model P23Gb), the pressures and blood flow rates were recorded on an ultraviolet light recorder (Honeywell, Visicorder 1508). Mean pressure was obtained by electrical integration and estimated by drawing a line by eye through the average pressure for the measurement period. In instances where the mean pressure was varying considerably it was estimated by using a planimeter. Zero pressures were determined post mortem as the levels of the tips of the cannulae when free in air. Systemic arterial pressure, pressure in the pulmonary artery pouch and limb perfusion pressure were recorded on a four channel tape

recorder (Hewlett-Packard No. 1105). Recordings were made at slow tape speed (15/16 in/sec). To allow rapid scanning of the total record at a shorter time base the tapes were played at fast tape speed (15 in/sec).

A. Limb And Kidney Perfusion

Control pressure in the pulmonary artery pouch was adjusted to about the same as the perfusion pressure of the left lungs at the start of the experiment (average 18 cm H_2O). This pressure was maintained for 3 min and a record for measurement of variables was taken during the last 1 min of the period. Perfusion pressure in the pouch was increased in steps of 10 cm of water between the range of 0-80 cm H_2O, then in steps of 20 cm of water until a maximum of 120 cm H_2O was reached. The pressure was raised for 3 min, and again a record for measurement of variables was taken during the last 1 min of each period. Between each distension as well as at the beginning and the end of the experiment the pulmonary artery pouch pressure was maintained at the control level for three minutes; these intervals serve as control periods. The average pressure obtained from the control periods before and after each experimental period were compared with those of the experimental period. Heart rate changes were treated similarly.

The isolated pouch of the main pulmonary artery and its branches were distended with various pressures in

12 dogs. Recordings began approximately two hours after thoracotomy had been performed. In six dogs (dogs No. 1-6) the pulmonary artery pouch was perfused with blood at 37°C taken from the reservoir and the hind-limbs were perfused with constant arterial blood flow. In another six dogs (dogs No. 7-12) the pulmonary artery pouch and the kidneys were perfused with blood at 37°C. The kidneys were perfused either with constant pressure or constant flow. Pouch perfusion was handled the same way as the limb perfusion experiments.

In four of the above dogs (dogs No. 5, 6, 11 & 12) the pulmonary artery was cooled down to different temperatures after completion of the above protocol. Cooling was achieved by infusing normal saline of 15°C and 30°C into the pulmonary artery pouch. Each cooling period was 3 minutes long and before and after each cooling period 37°C saline was used to bring the artery back to body temperature.

B. Cooling

In ten dogs the combined effect of cooling and distension of the pulmonary artery pouch was investigated. The pulmonary artery pouch was perfused with saline, the temperature of which could be varied. Control periods were taken with the main pulmonary arterial pouch pressure kept between 0 and 2 cm H_2O and the saline was just barely flow-

ing, allowing the temperature within the pouch to equilibrate to body temperature. Each control period was four minutes long with the variables measured during the fourth minute. Perfusion pressure was raised by increasing the saline flow to the pouch and adjusting the screw clamp on the drainage tubing. In each case when the perfusion pressure was raised a relatively large saline flow compared with the control was maintained, thus the temperature in the pouch was expected to become close to that of the saline, usually 30°C. The pressure was raised for three minutes and variables were recorded during the third minute. Approximately the same saline flow rate was used for each distension. Perfusion pressure was raised in steps of 10 or 20 cm H_2O in a similar fashion as before until a maximum of 120 cm H_2O was reached.

In eight of the dogs (dogs No.13, 14, 17-22) the vagus nerves were cut in the neck after completion of the above procedures and the experiments repeated. Before each experiment was repeated, the ability of the preparation to respond reflexly was checked by occluding both carotid arteries. In every case there was an increase in mean arterial pressure and an increase in heart rate.

C. Urine Collection

The ureters of the intact kidneys were cannulated in twenty-nine dogs for urine collection. Recordings began at least thirty minutes after the completion of the surgical

procedures. Each collection period was ten minutes long, the urine produced during that entire period was collected for analysis, a record of the different pressures was taken during the last 1 minute of each period.

For dogs numbering 23-32 the pulmonary artery pouch was perfused with venous blood at 37°C. The pressure was kept constant at 15 cm H_2O for 110 min. In these experiments the pulmonary arterial pouch pressure was kept constant, any changes in the urine composition and output are due to surgical interferences or just natural variation of the preparations. Since the next two sets of experiments were prepared in exactly the same fashion as dogs numbering 23-32, with the exception of the pulmonary arterial pouch pressures being varied, these animal thus serve as a set of control experiments which will account for time course changes not associated with the pulmonary arterial pouch pressure manipulations.

In nine dogs (dog No. 33-41) the pulmonary arterial pouch pressure was maintained at 5 cm H_2O for 40 min. Then the pressure was raised to 40 cm H_2O and kept at that level for 30 min. The pressure was lowered afterward back to 5 cm H_2O and maintained for another 40 min. Urine was collected as before. Blood pressures were recorded and urine samples were analysed.

For dogs numbered 42-51 the pulmonary arterial pouch pressure was kept at 15 cm H_2O for 40 min then incre-

ased to 80 cm H_2O for 30 min and released back to 15 cm H_2O for another 40 min. In order to maintain the glomerular ultra filtrate at a relatively constant composition (i.e. the blood composition), no extra fluid of any kind was added to the animals in these urine collection experiments other than priming the reservoir and the continuous infusion of the anesthetic agent.

D. Comparison Of Intact And Isolated Kidneys

The right kidneys of dogs numbered 52-57 were intact and the left kidneys were isolated and perfused with constant pressure throughout the experiment. The perfusion pump was adjusted such that the perfusion pressure was about the same as the systemic arterial pressure at the beginning of the experiment and kept constant by a negative feedback circuit. Pulmonary arterial pouch pressure in these animals maintained at 10 cm H_2O for 40 min, 100 cm H_2O for the next 30 min then lowered back to 10 cm H_2O for the last 40 min. Urine samples of the two kidneys were collected and analysed separately. The results were compared.

EVALUATION OF EXPERIMENTAL TECHNIQUES

A. Constant Flow/Pressure Preparation

By Ohm's law the relationship between the voltage, electric current and resistance in a simple resistive circuit can be expressed by

$$V = IR.$$

V = voltage drop across the resistor.

I = electric current through the resistor.

R = resistance.

The variables can be substituted and made to represent the blood pressure, flow and resistance across a cardiovascular bed as

$$P = FR.$$

P = pressure drop across a cardiovascular bed.

F = blood flow through the same cardiovascular bed.

R = resistance of the bed.

Suppose we want to measure the resistances of the cardiovascular bed, Ohm's equation can be rearranged to

$$R = \frac{P}{F}.$$

Since R is dependent of both P and F, both variables have to be measured to obtain R.

If we make one of the variables constant such as keeping the blood flow F constant the change in the resistance will be reflected directly by the other variable, in this case the perfusion pressure

$$R = \frac{\Delta P}{F}$$

also $$R \propto P.$$

To measure the change in the cardiovascular bed resistance will become a simple task of monitoring the change in the perfusion pressure. Similarily if the perfusion pressure P is kept constant, the change in resistance becomes

$$R \propto \frac{1}{F}.$$

All the experiments described in this thesis were done with a right heart bypass such that the venous blood was drained directly into the reservoir which is open to the atmosphere. Therefore we can consider the venous pressure as zero and pressure drop across any cardiovascular bed is the same as the arterial blood pressure it is subjected to.

For the limb perfusion experiments only constant flow technique was used to perfuse the hind-limbs. In the kidney perfusion experiments, because of the nature of the kidney, both constant flow and constant pressure methods were used to perfuse the kidneys in separate experiments.

A kidney is invested in a firm, strong fibrous capsule which does not stretch much even under high internal hydrostatic pressure. It is capable of autoregulating blood flow, it possesses a high oxygen consumption per unit of tissue weight and because of this, also requires a high blood flow. These properties make it necessary to utilize both methods to investigate kidney resistance changes.

With constant renal blood flow an increased renal resistance will cause vasoconstriction and elevate the interstitial pressure inside the kidney capsule. When the rise in interstitial pressure is excessive, it will compress the kidney tubules and arteries, increasing the renal resistance further by a mechanical means, as a result, highly augmenting the renal pressure. With constant pressure, on the other hand, increased kidney resistance also causes vasoconstriction. Since the renal pressure is constant, the constriction is reflected by a decreased renal blood flow. The constriction can become so severe that renal blood flow can virtually cease. Since the kidney requires a large amount of blood supply for its oxygen consumption, excessively low renal blood flow for a prolonged time can cause tissue hypoxia and permanently damage the kidney. Therefore constant pressure and constant flow methods were used to perfuse the kidneys hoping to eliminate the undesirable effects of both and permitting us to observe the true changes in the kidney resistance.

B. Heart Rate Calculation

Electrocardiogram was recorded from leads placed in the left chest and the right leg region. Heart rate was calculated by counting the electrocardiogram over a period of 30 sec and extrapolated to beats per minute. At the end of this process of extrapolation a possible error of ± 2 beats per minute has been introduced into the heart rate.

Any changes of less than ± 2 beats per minute should thus not be considered as real changes.

C. Pressure And Flow Recordings

The pressures were recorded by using strain gauge transducers, after proper amplification they are displayed on the ultra-violet light recorder. At the beginning of each experiment the pressure recording system was calibrated against a mercury manometer or a saline column over several fixed values. Display scale of 10 cm H_2O/cm was used for the pulmonary arterial pouch pressure, 10 mm Hg/cm was used for the arterial, limb and kidney pressures. For the constant pressure kidney perfusion experiments the kidney pressure scale used was 20 mm Hg/cm. In each case the resolution of the record was 0.5 mm, thus ±0.5 cm H_2O, ±0.5 mm Hg, and ±1.0 mm Hg for the three different scales. Cardiac output, limb and kidney perfusion blood flows were monitored with pulsed logic electromagnetic flow meters, and after proper amplifications displayed on the ultra-violet recorder for measurement. The performance of the flow meters were tested in steps between the flow rate of 50 and 3500 ml/min. Results show that in the tested range the flow meter output is linear. Display scale of about 200 ml/min/cm was used for the cardiac output measurement. Limb and kidney blood flow was measured on a scale of about 25 ml/min /cm. No significant drift was observed in either the pressure and flow recording systems over the entire experi-

mental period.

D. Urine And Plasma Analysis

Urine and plasma were analysed for sodium and potassium concentration by using a flame photometer after dilution. The flame photometer utilises a lithium standard (15 mEq Li/L). The emission intensities of sodium and potassium were compared with that of the lithium standard and a built-in analog computer provided a digital readout via a servo-counter.

The machine was calibrated with standard solutions. A solution containing 100 mEq/L of sodium and 100 mEq/L of potassium was used as a standard when urine samples were analysed. A solution containing 140 mEq/L of sodium and 5.0 mEq /L of potassium was used when plasma samples were analysed. The flame photometer was recalibrated after every 5 to 10 samples. Generally the readjustment necessary when recalibrating with the standards was 1 or 2 mEq/L for the urine and 0.1 mEq/L for the plasma sodium standard. Duplicate samples were used to check the total error introduced by the diluter and the flame photometer, in ten tests the error was found to be within ± 4%.

Plasma and urine osmolarity was measured using the method of freezing point depression. The osmometer utilizes the supercooling technique and the sample was induced to freeze by stirring. A thermistor probe measured the temp-

erature of the sample throughout the entire process. Calibration of the osmometer was checked with standard solutions of 100 mOs/kg and 500 mOs/kg at the beginning of each sample batch. The osmometer was accepted as calibrated if the reading of the standards were within 5 mOs/kg of the manufacturer's value. Testing with seven duplicate samples showed a possible error of 5% in the osmolarity reading.

Urine volumes were measured in graduated cylinders at the time the urine was collected. If the urine volume for the 10 min collection period was less than 10 ml, the urine was measured in 10 ml graduated cylinders, the accuracy of which is ± 0.05 ml. If the urine volume exceeded 10 ml in 10 min the urine was measured in 25 ml graduated cylinders, the accuracy of which is ± 0.1 ml. Sodium and potassium excretions are calculated by multiplying the sodium and potassium concentrations with the urine volume.

Osmolar clearance is obtained by using the equation

$$C_{os} = \frac{U_{os}}{P_{os}} \times V$$

C_{os} = osmolar clearance

U_{os} = urine osmolarity

P_{os} = plasma osmolarity

V = urine volume

C_{H_2O} = free water clearance

and free water clearance is calculated from

$$C_{H_2O} = V - C_{os}.$$

E. Feedback Control Circuits

Constant pressure or constant blood flow perfusion was made possible by electric feedback control circuits. These circuits monitor the performance of the system and make appropriate adjustments, if necessary, to maintain constant perfusion pressure or flow. The circuit consists of a sensor (pressure transducer or flow meter) which senses the controlled variable (blood pressure or blood flow) constantly. Signal from the sensor after proper amplification is fed into a comparator which compares the sensor signal with a pre-set standard signal. The comparator acts as a subtractor which subtracts the sensor signal from the standard signal and the output of the comparator is the difference of the two. Again after proper amplification the comparator output is used to control the roller pump (Fig. 4). Any deviation of the sensor signal from the standard signal will cause the pump to change its speed and/or flow direction thus bringing the pressure or flow rate back to the pre-set standard level. Different values of the controlled variable may be set by alteration of the standard signal.

The feedback control circuits were highly effective. In all the experiments reported there was no measurable error credited to the control system. With a resolution of 0.5 mm of the ultra-violet light recorder there was no noticeable fluctuation of the controlled variable throughout the entire perfusion period which could be as long as

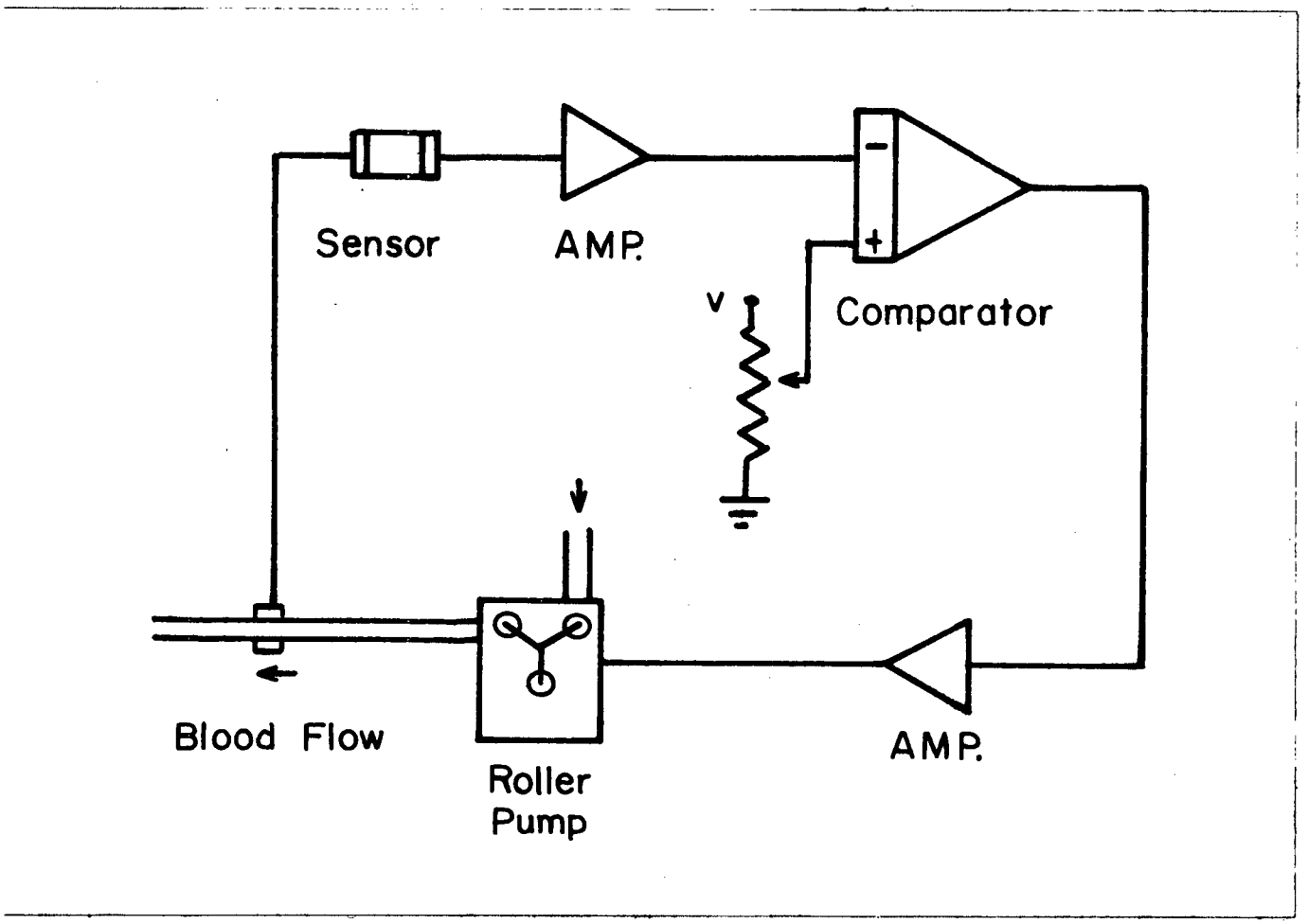

Fig. 4 Feedback circuit for the roller pump control. Same circuit was used in constant pressure and constant flow perfusion.

4½ hours.

F. Mathematical Methods

a) Correction to absolute zero pressure

Systemic arterial pressure changes in response to the pulmonary arterial pressures for each individual experiments, were rearranged first, before averaging for the series. During the experiment the absolute pressure at the tip of the pulmonary artery pouch cannula was not known, zero pressure was taken at a fixed reference. At the end of the experiment the cannula was freed in the air and the pressure difference between the tip of the cannula and the reference was measured and correction was made such that the pressure at the freed cannula tip was considered as the absolute zero. Raw data were plotted on graph papers, straight lines were drawn between neighbouring points then zero correction was made. Stepwise increments of 10 cm H_2O in the pulmonary arterial pouch were marked using the absolute zero as reference. The changes in systemic pressure at pressures of 10, 20, 30, 40, 50, 60, 70, 80, 100, and 120 cm H_2O in the pulmonary arterial pouch were extrapolated from the graph and averaged between individual experiments of the same set.

b) t-test

When paired results were analysed the t-test for paired values was used. In each case the value obtained

during the experimental period was compared with the mean value of the two control periods before and after it. Differences were considered significant if $2P<0.10$ or $P<0.05$. The choice of one or two tailed test was selected for appropriate testing conditions.

c) Wilcoxon two sample rank test (Mann-Whitney U-test)

Results from the urine collection experiments were tested with nonparametric statistical method. Each of the urine collection experiments was divided into three sections. The first forty-minute section is called period I, the second thirty-minute section is called period II, and the last forty-minute section is called period III.

Period I	Period II	Period III

Since each variable was measured once every ten minutes, each period contains either three or four samples of the same variable. In the control experiments, the pulmonary artery pouch pressure was constant throughout periods I, II and III. In two other sets of experiments the pulmonary artery pouch pressure was either at 5 or 15 cm H_2O during periods I and III and was raised to 40 or 80 cm H_2O during period II. The sample values of each variable within each period was then averaged and compared with that of another period. As a result, a set of ten numbers was obtained from the ten control experiments by subtracting the average sodium excretion rate in period I from that of period

II. Similarily, a set of nine numbers were obtained from the nine experiments where the pulmonary artery pouch pressures were raised to 40 cm H_2O in period II. These two sets of 10 and 9 numbers are tested by the Wilcoxon two-sample rank test (Goldstein, 1968). Any significant difference between the two sets will be the result of the increase in the pulmonary artery pouch pressure. Natural variations caused by the surgical maneuver will be eliminated.

RESULTS

A. Cardiovascular Effects Of Graded Pulmonary Artery Distension

a) Systemic pressure and heart rate changes

There were no differences in the changes in systemic pressure in response to changing the pressure in the isolated pulmonary artery pouch between the six dogs in which the hind-limbs were perfused and the six dogs in which the kidneys were perfused and therefore these results have been pooled. Fig. 5 shows the average results of 14 tests (in two dogs two sets of pouch distensions were performed) in which venous blood at 37°C was used for the pulmonary artery perfusion.

Mean cardiac output of the experiments was 1820 ml/min, ranging from 755 to 2240 ml/min. The average control pouch pressure was 18 cm H_2O (range 10 to 24 cm H_2O), and that of the systemic pressure was 124 mm Hg (range 104 mm Hg to 149 mm Hg). Increase of the pulmonary artery pouch pressure caused a rise in the systemic pressure proportional to the rise in the pulmonary arterial pressure. The change was small with pouch pressure between 10-30 cm H_2O, but became prominent beyond that. At pouch pressure of 120 cm H_2O, the maximum tested, the average increase in systemic pressure was 12 mm Hg, 10% of control. Paired t-test was used to examine the changes which occurred by raising the

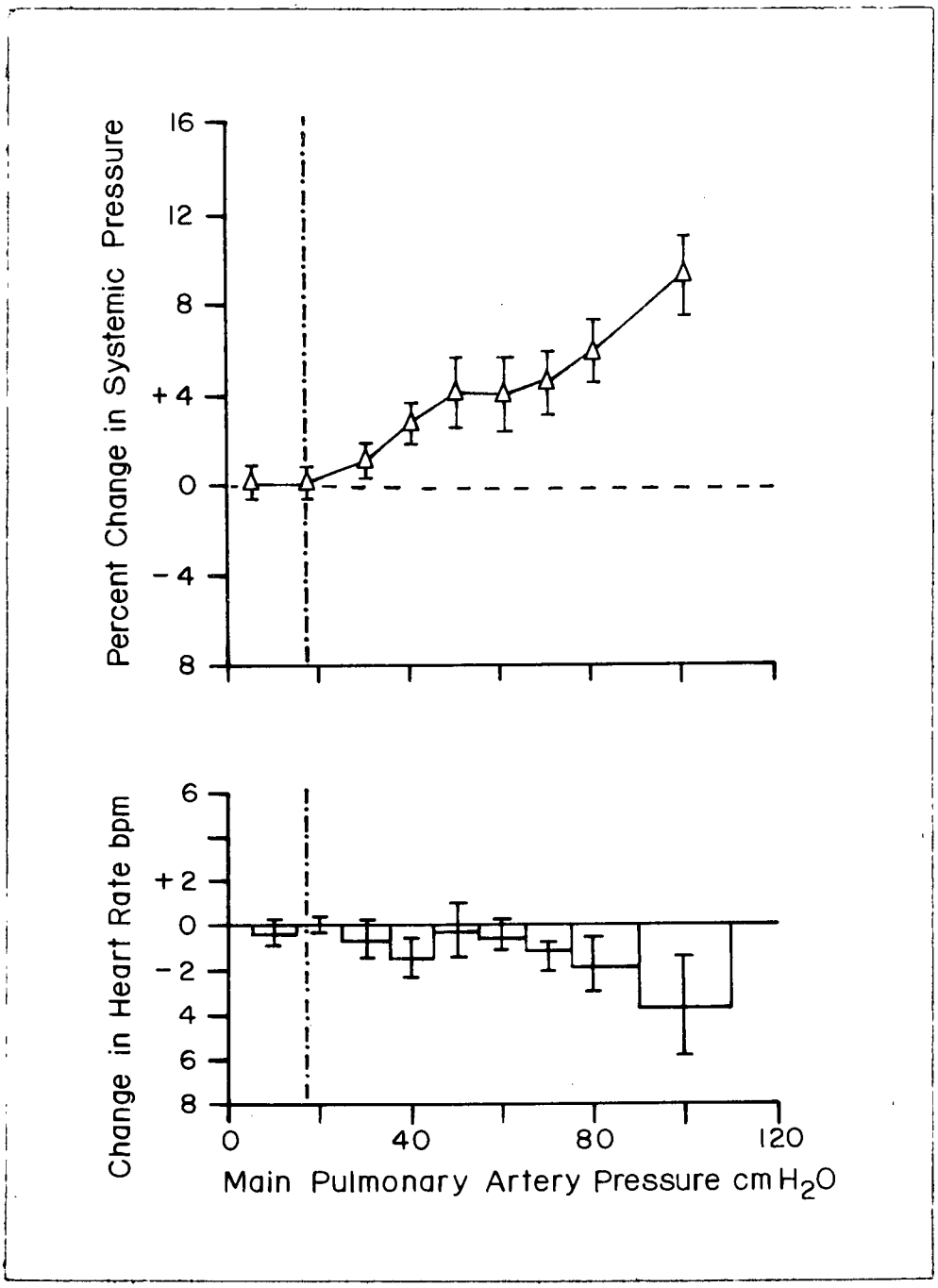

Fig. 5 Changes in systemic pressure and heart rate caused by varying the pulmonary arterial pouch perfusion pressure between 5 and 120 cm H_2O. Values plotted are changes from control values observed with the pulmonary arterial pouch pressure maintained at 18 cm H_2O. Values shown are the averages in 12 experiments $\pm$ SEM.

pouch pressure from control to 40 cm H_2O which is considered a physiological pressure. This maneuver resulted in a 2% rise of the systemic pressure corresponding to about 3 mm Hg (Student's t-test; $2P<0.005$). Therefore within the physiological range although the increase in systemic pressure was small, it was highly significant. From Fig. 5 it is quite obvious that any further increment of the pouch pressure beyond 40 cm H_2O will also cause a statistically significant rise in the systemic pressure. The systemic pressure changes reflect the changes in general peripheral resistance because the cardiac outputs were kept constant in these experiments.

Average control heart rate in these experiments was 143 beats/min (range 92 to 183 beats/min). The mean response to pulmonary arterial pouch pressure increase appeared to be a slight decrease in heart rate when pulmonary arterial pouch pressure was high (Fig. 5). However, the changes were always small and usually within ± 2 beats/min, which is the range of error of counting over 30 sec periods. Paired t-test of heart rate during the control and experimental periods with pulmonary arterial pouch pressure of 100 cm H_2O showed no significant difference ($2P>0.1$).

b) Change in limb resistance

Hind-limb vascular resistance was measured in six dogs and was calculated by dividing the perfusion pressure of the hind-limb by the blood flow. Since the hind-limb

blood flow was constant and the right atrium was connected to the reservoir, which was open to the atmosphere maintaining outflow pressure constant, the change in limb resistance was directly proportional to the change in the limb perfusion pressure.

Fig. 6 shows that when the pulmonary artery pouch pressure was raised above the control value (average 18 cm H_2O) limb resistance increased in a proportional fashion. As the pouch pressure was decreased there was a slight drop in the average limb resistance. Mean control limb pressure was 99 mm Hg and ranged from 82 to 108 mm Hg. Average limb blood flow was 182 ml/min ranging from 125 ml/min to 285 ml /min. At pouch pressure of 100 cm H_2O the average rise in limb resistance (pressure) was 17% in contrast with only 8% in the systemic pressure. Compared with the increase in the systemic arterial pressure, the limb vascular resistance almost always showed a greater percentage increase at any pulmonary artery pouch pressure.

Fig. 7 is part of the record of an experiment recorded on tape and played back at a reduced time base. Graded responses of the systemic arterial pressure and limb pressure are clearly seen in response to different pulmonary arterial pouch pressures in the range of 50-80 cm H_2O. It may also be seen that peak responses are observed about 1 minute after the change in pulmonary arterial pouch pressure and that there has been some reduction in response to a

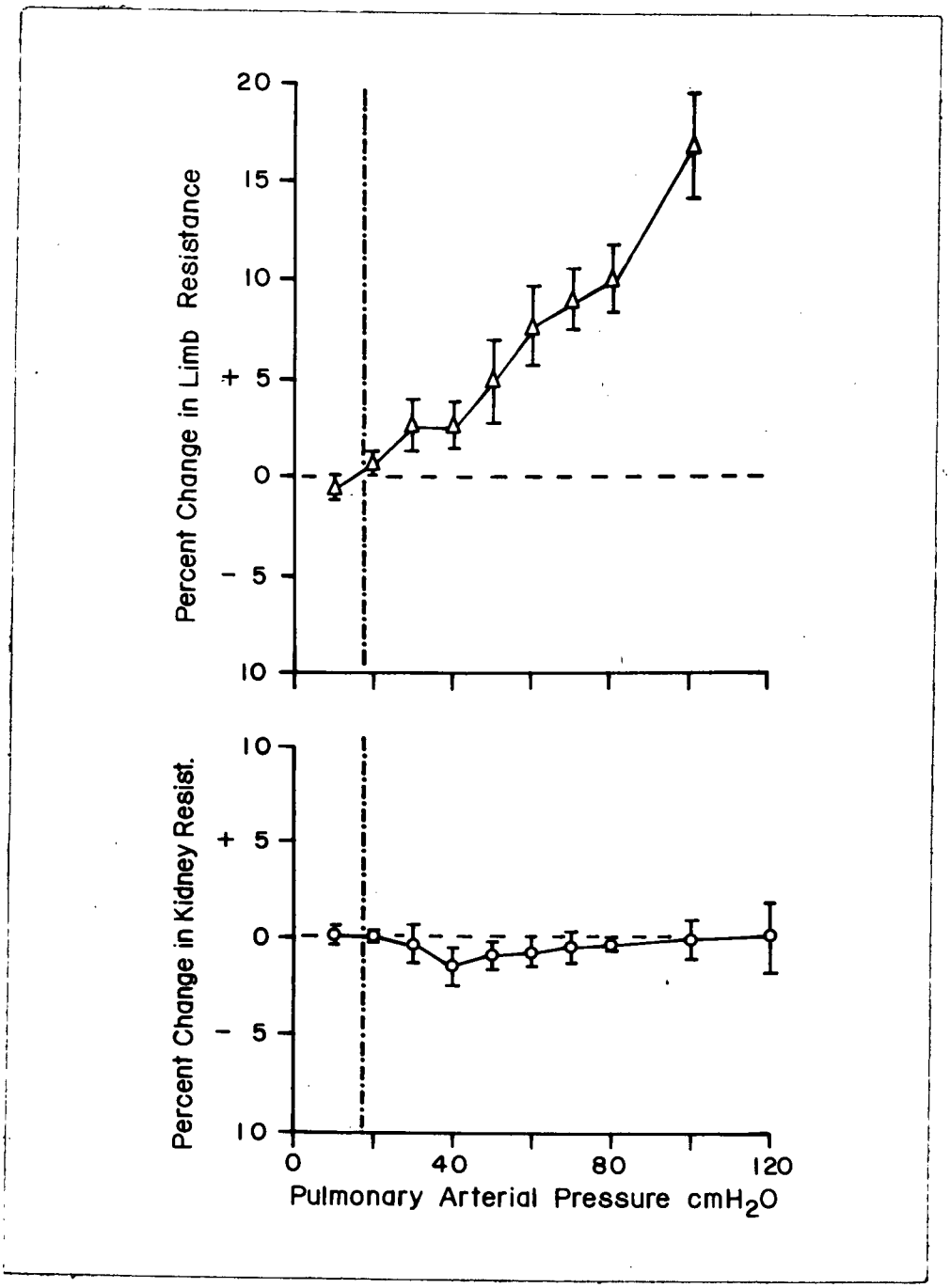

Fig. 6 Changes in hind-limb and renal resistance in response to changes in perfusion pressure in a pulmonary arterial pouch. Results for limb resistance are average changes from values at a pulmonary arterial pouch pressure of 18 cm H_2O in 6 dogs $\pm$ SEM. Results for renal resistance are calculated similarily in 8 tests in 6 dogs.

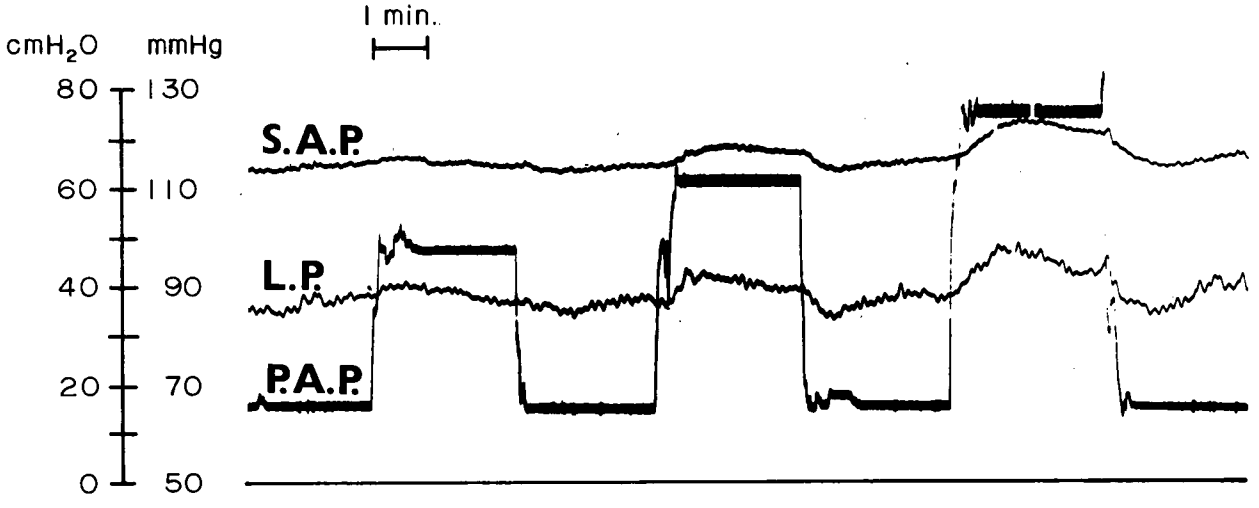

Fig. 7 Part of the record of one experiment showing changes in systemic arterial pressure (SAP, mm Hg), hind-limb perfusion pressure (LP, mm Hg) and pulmonary arterial pouch pressure (PAP, cm H_2O). Time base has been reduced by recording on tape at slow speed and replaying at a higher speed. Graded responses are shown to increasing pulmonary arterial pouch pressure over the range 45-75 cm H_2O.

more steady state by the third minute when measurements were made. In fact the transient changes were more rapid and somewhat greater than appears from this record as the combination of the reduced time base and electronic damping of the systemic pressure and limb perfusion pressure traces increases the apparent time constant of the system. The pulmonary arterial perfusion trace was undamped and shows that the oscillations imposed on the system by the roller pump were usually less than 5 cm H_2O in amplitude.

c) Changes in kidney vascular resistance

Six dogs were prepared for kidney perfusion. In two of the animals, two sets of pouch distensions were performed, thus a total of eight sets of results were avaliable. Three tests were carried out with constant renal blood flow perfusion and five tests were done with constant renal arterial blood pressure. The change in renal vascular resistance was measured as either the change in renal blood flow or the change in renal perfusion pressure. As the results were similar in each case they have been pooled.

Fig. 6 shows the average results of the eight tests. The change in renal resistance in response to changes in pulmonary arterial pouch pressure were small and inconsistent. Maximum average change was -1.5% compared with 17% in the limbs. In contrast to the limb resistance the renal vascular resistance is not significantly affected by

changes of the pouch pressure. The average control arterial pressure was 105 mm Hg with a minimum of 83 mm Hg and a maximum of 125 mm Hg. Since in some cases only one kidney was perfused and in others both kidneys were perfused, no comparison of renal blood flow is made between individual dogs. In all experiments the renal blood flows were found to be of the order of 1-3 ml/min/gm kidney perfused.

d) Perfusion with saline at different temperatures

In almost all of the experiments reported above, the preparation remained stable and responsive at the completion of the procedures. Saline, at different temperatures, was perfused into the pulmonary arterial pouch to investigate its response to cooling. Fig. 8 is a reduced time base playback of one typical experiment with sections of cooling and rewarming. Decrease in systemic arterial pressure and limb perfusion pressure were obvious in every test when the temperature of the saline was changed from 37°C to 30°C or to 15°C. This effect was present at either low (15 cm H_2O) or high (80 cm H_2O) pulmonary artery pouch pressure. In both cases the decrease in systemic and limb pressure was more than 10 mm Hg. No quantitative analysis of these response has been made because of limitation of equipment to give adquate temperature control of the saline. Instead another series of experiments was performed which would illustrate the combined effect of cooling and distending the pulmonary arterial pouch.

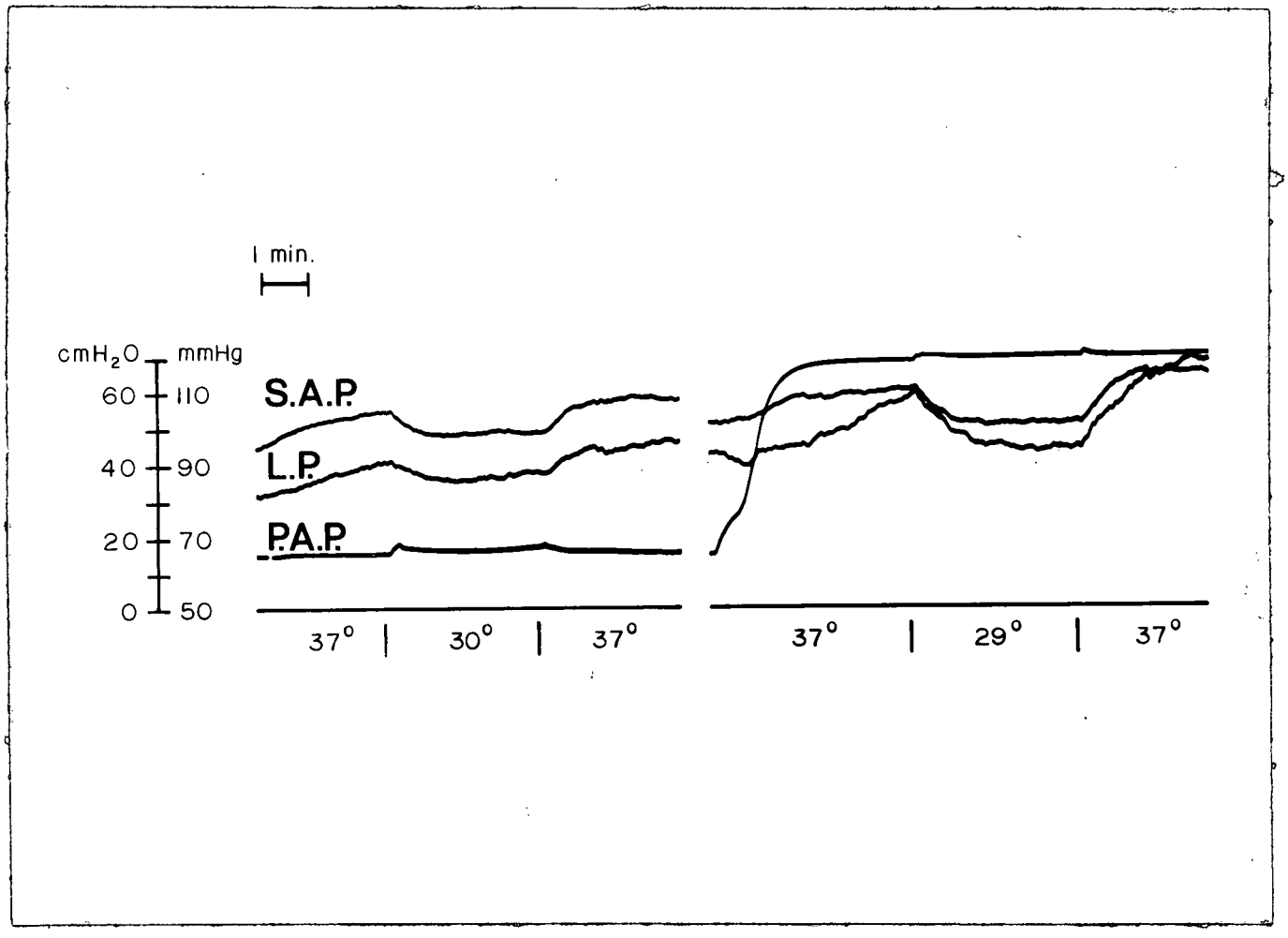

Fig. 8 Parts of the record of one experiment showing changes in systemic arterial pressure (SAP, mm Hg) and hind-limb perfusion pressure (LP, mm Hg) in response to changing the temperature in the pulmonary arterial pouch. Records taken at low and high pulmonary arterial pouch pressure (PAP, cm H_2O). The temperature in the pouch immediately before the first panel has been 15°C.

B. Distension Of The Pulmonary Artery With 30°C Saline

In ten dogs experiments were performed to examine the interaction of cooling and pulmonary arterial pouch pressure. The pulmonary artery pouch was perfused with saline at 30°C. During the control periods saline flow was extremely slow (<5 ml/min) and pressure was low (0-2 cm H_2O), during the experimental periods when the pressure was raised the flow was rapid (approximately 100 ml/min), thus exerting a cooling effect on the pulmonary arterial wall. The precise temperature at the receptor site is unknown but believed to be less than 37°C.

Fig. 9 shows the change in systemic pressure and heart rate observed during combined cooling and increasing pressure in the pulmonary artery pouch. There was a reduction in systemic pressure when pouch pressures were low (10-60 cm H_2O). Distension with higher pressure (60-120 cm H_2O) caused an increase in systemic pressure despite the cooling. Mean control systemic pressure was 117 mm Hg, ranged from 93 to 139 mm Hg. Maximum average drop in systemic pressure was 7 mm Hg when the pouch pressure was at 10 cm H_2O. Maximum mean increase in systemic pressure was 10 mm Hg, and occurred at a pouch pressure of 120 cm H_2O, which was the highest pouch pressure tested. There were no net changes in the systemic pressure at the pouch pressure of 60 cm H_2O; presumably the cooling and the distension effects cancelled each other out. Comparison with Fig. 5 indicates that cool-

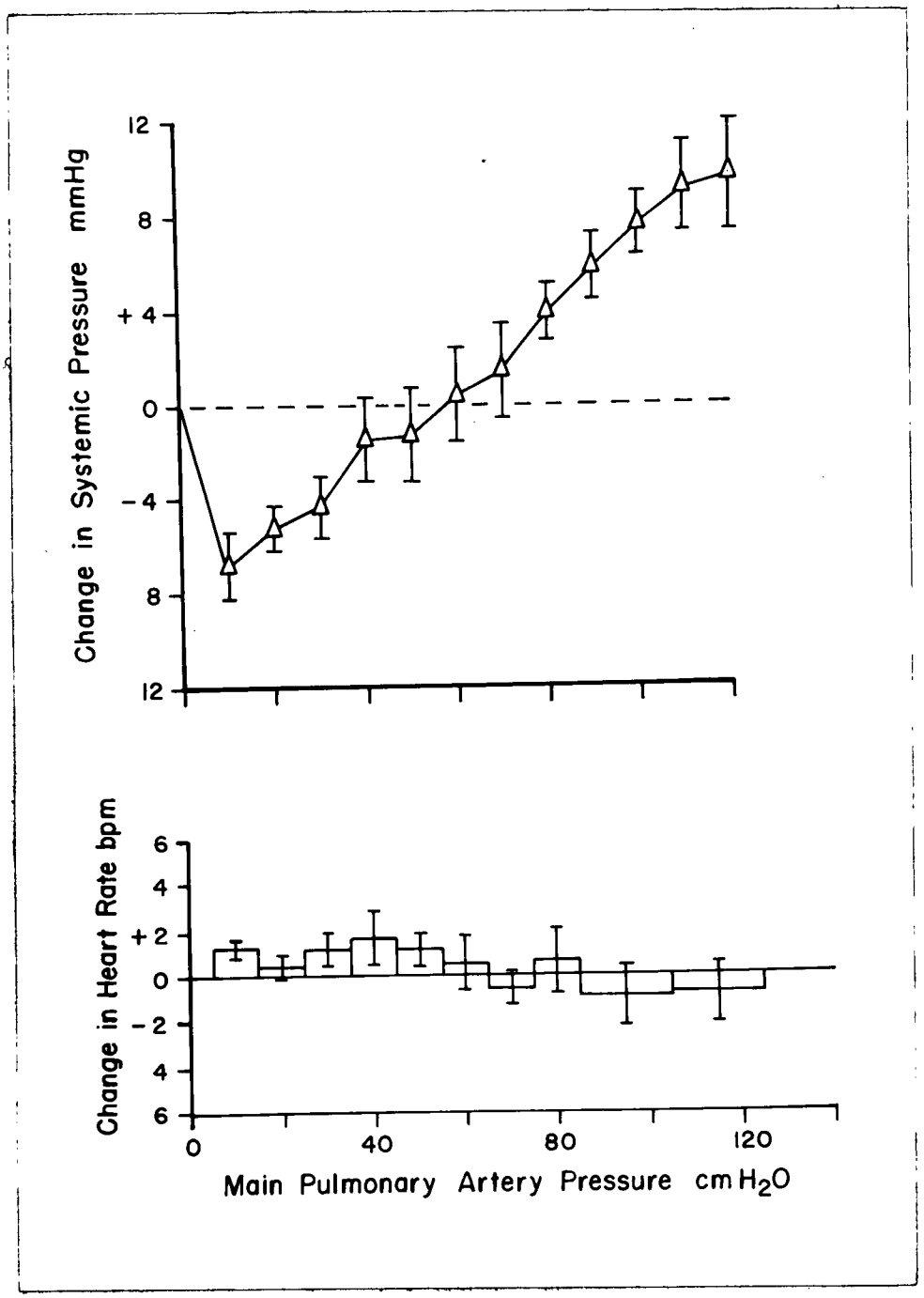

Fig. 9 Changes in mean systemic arterial pressure and heart rate associated with increasing pouch pressure from 0-2 cm H_2O and simultaneously cooling the pulmonary arterial pouch from 37°C to 30°C. Average results (± SEM) in 10 dogs.

ing to 30°C may have been somewhat more effective at reducing systemic pressure at low pulmonary arterial pouch pressure than at high pouch pressure. Mean control heart rate was 152 beats/min, and average changes in response to the pouch pressure were always small and within ±2 beats/min as shown on Fig. 9.

C. Effect Of Vagal Section

In eight of the ten animals tested above, the vagus nerves were cut in the neck, and the pulmonary artery pouch was again perfused with 30°C saline over the whole pressure range. Any reflex response from the pulmonary artery which requires the vagus nerves as part of the pathway will be eliminated by this procedure.

After vagotomy, the average control systemic pressure was 107 mm Hg with minimum at 94 mm Hg and maximum at 120 mm Hg. The average control heart rate was 165 beats/min. Variation of the pulmonary arterial pouch pressure between 10 and 120 cm H_2O caused no significant changes in the systemic pressure. The maximum average change was -1 mm Hg in contrast with 10 mm Hg when the vagus nerves were intact and 12 mm Hg when warm blood at 37°C was used. Fig. 10 shows that the change in systemic pressure vs. pouch pressure curve is virtually parallel to the abscissa. Comparison with Fig. 9 will show readily that the vagus nerves are essential for both the cooling and distension reflex from

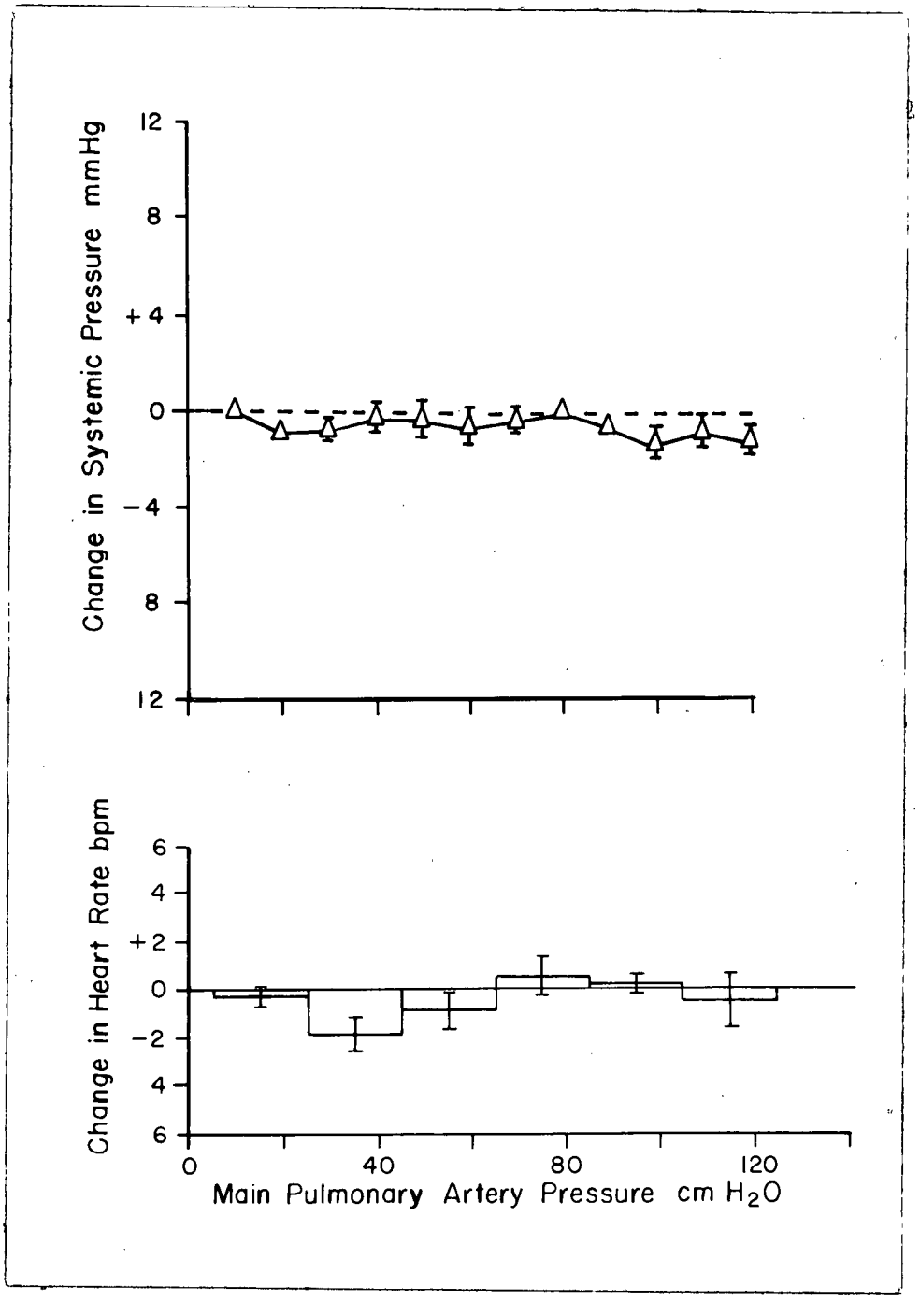

Fig. 10 Repetition of the experiment shown in Fig. 9 after cutting both cervical vagus nerves. Average results (±SEM) from 8 of the 10 dogs used in Fig. 9.

the pulmonary artery. Heart rate changes after vagotomy again were always within the range of counting error.

D. Urine Collection Experiments

To collect urine samples for analysis a volume of at least 1 ml is needed, therefore ten minutes was allowed for each collection period. As has been outlined in the methods section, every experiment required 110 min to complete, during this long procedure, changes in both hemodynamic and renal functions are possible. In ten animals all surgical procedures were completed,but pulmonary arterial pouch pressure was maintained constant. These served as controls to give an indication of the degree of change and provide as a basis for comparison.

a) Cardiovascular effects

Distension of the pulmonary artery pouch with blood at 37°C has been shown to cause an increase in the systemic pressure. Results shown on the graphs in the previous section (Fig. 5) were taken at the third minute of a three minute period of distension. In the urine collection experiments the pulmonary arterial pouch was distended for thirty minutes. Changes observed in these experiments will exclude any transient response which might occur, and the results represent steady state reflex changes initiated by distension of the pouch of the pul-

monary artery. The pouch pressures were maintained close to the desired pressures and very few minor adjustments were necessary during the course of the experiment.

Results from the control experiments are shown in Fig. 11, where the pouch pressure was kept constant at 15 cm H_2O for the 110 min experimental period. Average systemic pressure was 115 mm Hg. Except for small fluctuations, the systemic pressure was steady throughout the experiment. Fig. 12 represents the results of nine dogs in which the systemic pressure was 112 mm Hg at the beginning of the experiments where the pouch pressure was kept at 5 cm H_2O. Increasing the pouch pressure to 40 cm H_2O caused a small increase in systemic pressure of approximately 3-4 mm Hg. Fig. 13 represents the results on 10 dogs where the pulmonary arterial pouch pressure was raised from 15 cm H_2O to 80 cm H_2O, the systemic pressure increased from 128 mm Hg to 135 mm Hg during the time the pulmonary arterial pouch pressure was raised. The increase in systemic pressure was maintained during the entire test period of 30 min and when the pouch pressure was released back to 15 cm H_2O, the systemic pressure dropped back to about 128 mm Hg. This response was consistent and was seen in all ten animals tested.

The systemic pressure responses of the three sets of experiments are shown on Fig. 14 for comparison. The results are consistent with those shown in Fig. 5 in which

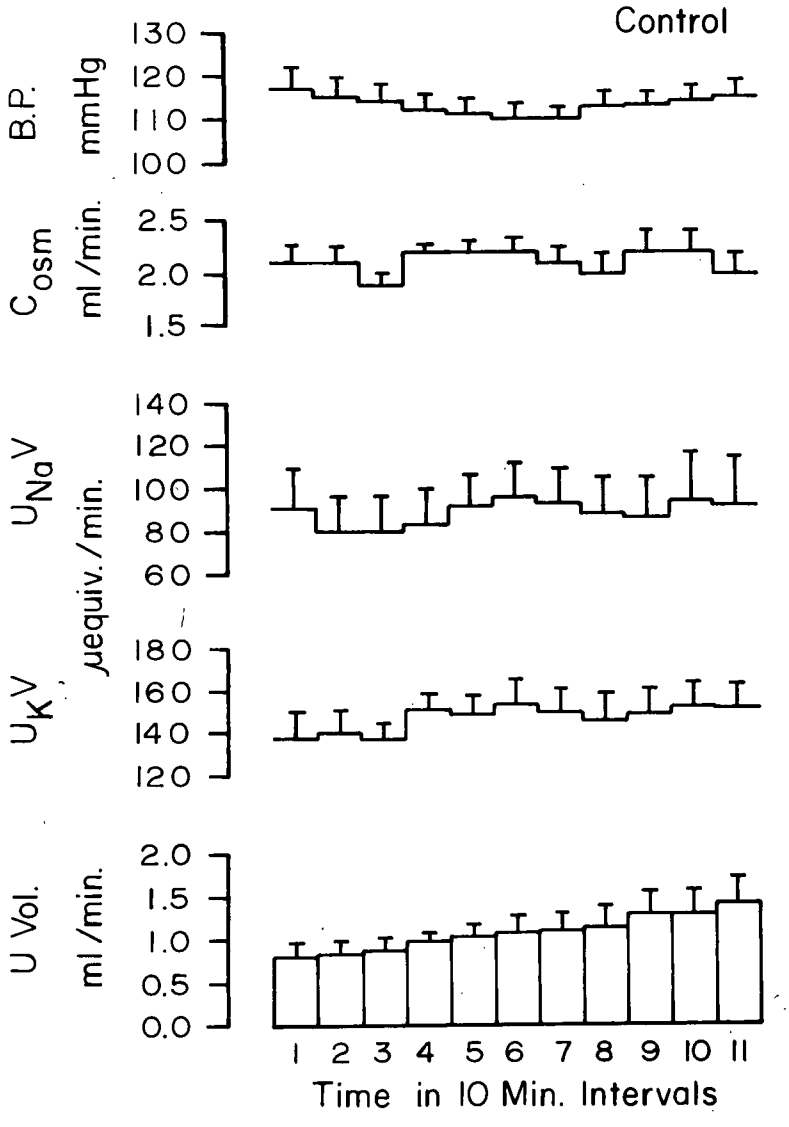

Fig. 11 Changes in systemic arterial pressure, urine output and composition associated with time. Pulmonary arterial pouch pressure was kept at 15 cm H_2O throughout the experiment. Average results (± SEM) in 10 dogs.

TABLE 2

Control experiments (urine collection)

Time 10 min periods	Sys P mm Hg $\bar{x} \pm$	SEM	C_{os} ml/min $\bar{x} \pm$	SEM	C_{H_2O} ml/min $\bar{x} \pm$	SEM	$U_{Na}V$ µEq/min $\bar{x} \pm$	SEM	U_KV µEq/min $\bar{x} \pm$	SEM	U_{vol} ml/min $\bar{x} \pm$	SEM
1	117	5	2.1	0.2	-1.3	0.1	91	19	138	12	0.8	0.2
2	115	4	2.1	0.2	-1.2	0.2	80	16	140	11	0.9	0.2
3	114	4	1.9	0.1	-1.1	0.2	76	17	137	8	0.9	0.2
4	112	3	2.2	0.1	-1.2	0.1	83	16	151	8	1.0	0.1
5	111	3	2.2	0.1	-1.1	0.1	92	15	149	10	1.1	0.1
6	110	3	2.2	0.1	-1.1	0.1	96	16	154	12	1.1	0.2
7	110	3	2.1	0.2	-1.0	0.2	93	15	150	12	1.1	0.2
8	112	3	2.0	0.2	-0.8	0.2	88	17	146	13	1.2	0.3
9	112	3	2.2	0.2	-1.0	0.4	86	19	149	13	1.3	0.3
10	114	4	2.2	0.2	-0.8	0.2	94	22	153	12	1.3	0.3
11	115	4	2.0	0.2	-0.5	0.3	92	22	152	11	1.4	0.3

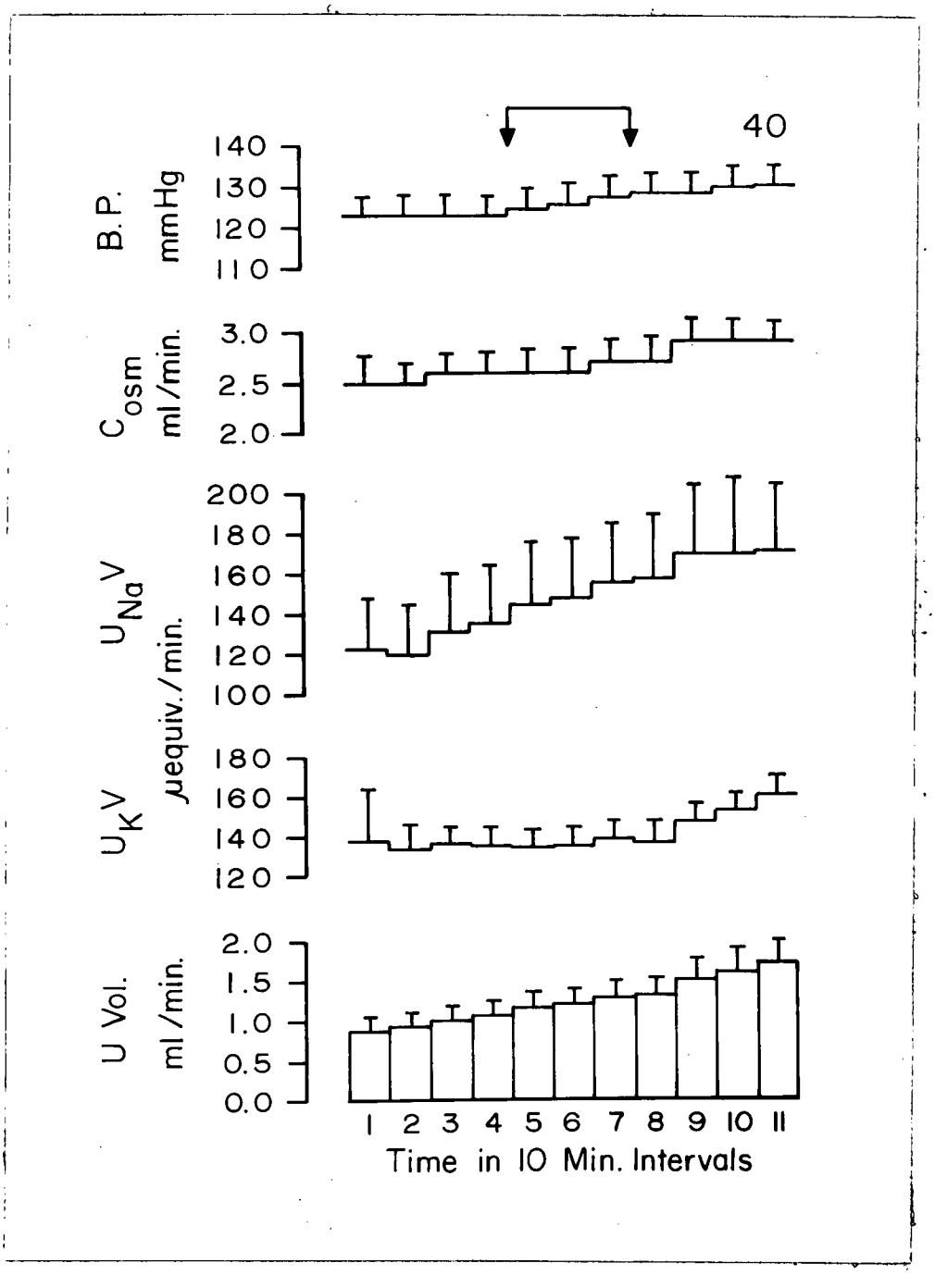

Fig. 12 Changes in systemic arterial pressure, urine output and composition in response to pulmonary arterial pouch distension to 40 cm H_2O (period between the arrows). During the control periods, before and after the distension pouch pressure was kept at 5 cm H_2O. Average results (± SEM) in 9 dogs.

TABLE 3

Pulmonary artery distension to 40 cm H_2O
(urine collection)

Time 10 min periods	Sys P mm Hg $\bar{x} \pm$ SEM		C_{os} ml/min $\bar{x} \pm$ SEM		C_{H_2O} ml/min $\bar{x} \pm$ SEM		$U_{Na}V$ µEq/min $\bar{x} \pm$ SEM		U_KV µEq/min $\bar{x} \pm$ SEM		U_{vol} ml/min $\bar{x} \pm$ SEM	
1	123	5	2.5	0.3	-1.6	0.2	123	25	137	21	0.9	0.2
2	123	5	2.5	0.2	-1.5	0.1	120	24	123	12	0.9	0.2
3	123	5	2.6	0.2	-1.6	0.1	132	27	136	9	1.0	0.2
4	123	5	2.6	0.2	-1.5	0.2	136	28	135	10	1.1	0.2
5	125	5	2.6	0.2	-1.5	0.2	145	30	134	9	1.2	0.2
6	126	5	2.6	0.2	-1.4	0.2	149	30	135	10	1.2	0.2
7	128	5	2.7	0.2	-1.4	0.2	156	29	138	9	1.3	0.2
8	129	5	2.7	0.3	-1.4	0.2	158	32	136	11	1.3	0.2
9	129	5	2.9	0.2	-1.4	0.3	171	34	147	10	1.5	0.3
10	131	5	2.9	0.2	-1.3	0.3	172	36	152	10	1.6	0.3
11	132	5	2.9	0.2	-1.2	0.3	172	32	160	10	1.7	0.3

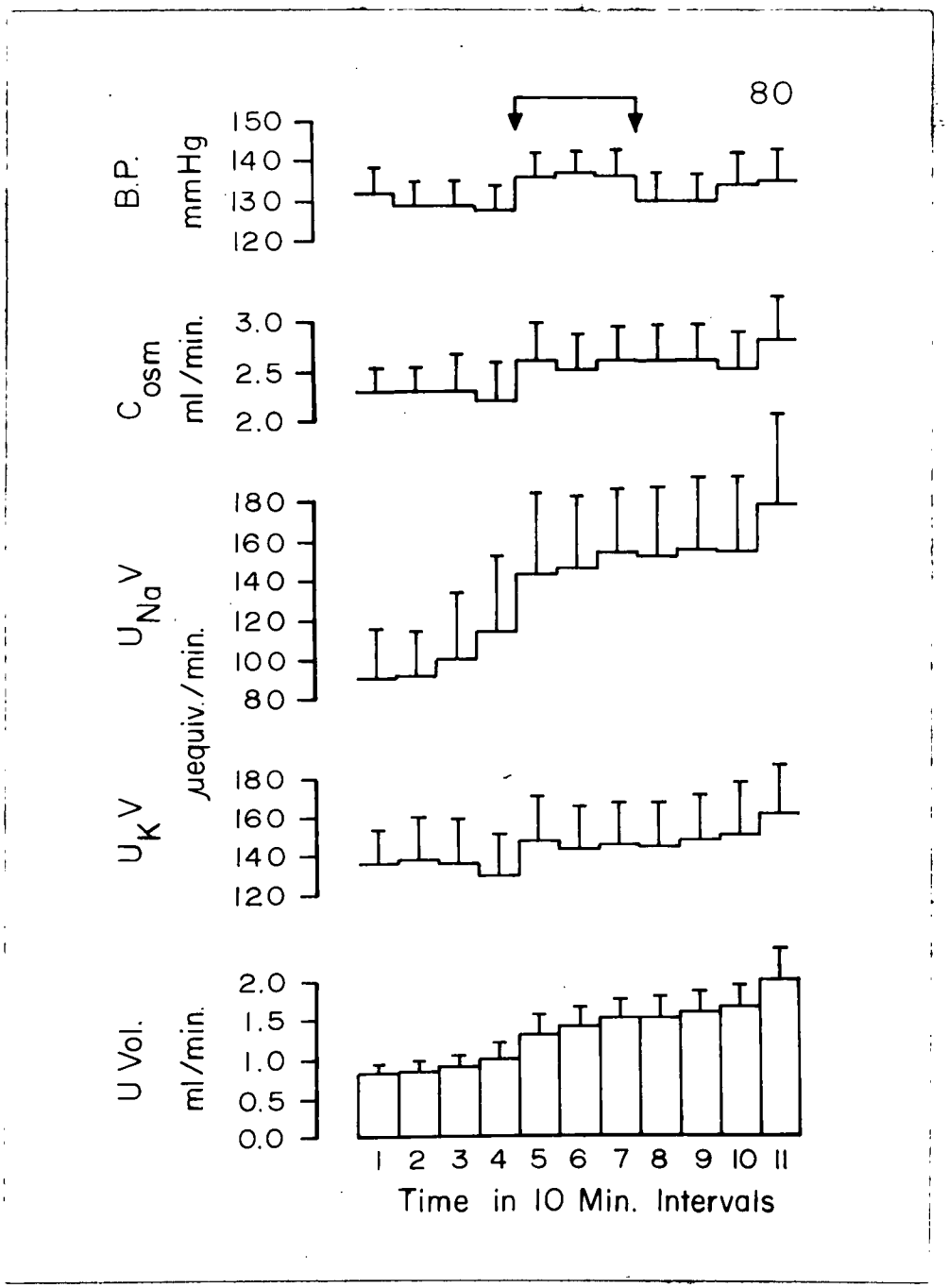

Fig. 13 Changes in systemic arterial pressure, urine output and composition in response to pulmonary arterial pouch distension to 80 cm H_2O (period between the arrows). During the control periods, before and after the distension, pouch pressure was kept at 15 cm H_2O. Average results (± SEM) in 10 dogs.

TABLE 4

Pulmonary artery distension to 80 cm H_2O

Time 10 min periods	Sys P mm Hg $\bar{x} \pm$ SEM	C_{os} ml/min $\bar{x} \pm$ SEM	C_{H_2O} ml/min $\bar{x} \pm$ SEM	$U_{Na}V$ µEq/min $\bar{x} \pm$ SEM	U_KV µEq/min $\bar{x} \pm$ SEM	U_{vol} ml/min $\bar{x} \pm$ SEM
1	132 6	2.3 0.3	-1.5 0.2	91 26	137 17	0.8 0.1
2	129 6	2.3 0.3	-1.4 0.2	91 24	138 20	0.9 0.1
3	129 6	2.3 0.4	-1.5 0.3	100 34	137 22	0.9 0.2
4	128 6	2.2 0.4	-1.3 0.2	114 40	130 21	1.0 0.2
5	136 6	2.6 0.4	-1.3 0.2	143 42	148 23	1.3 0.3
6	137 5	2.5 0.4	-1.1 0.2	146 37	144 22	1.4 0.3
7	136 6	2.6 0.3	-1.1 0.2	154 33	146 22	1.5 0.2
8	130 7	2.6 0.4	-1.1 0.2	152 36	145 22	1.5 0.3
9	130 7	2.6 0.4	-1.0 0.3	155 38	149 23	1.6 0.3
10	134 8	2.5 0.4	-0.9 0.3	154 39	151 21	1.6 0.3
11	135 8	2.8 0.4	-0.8 0.4	178 46	162 25	2.0 0.4

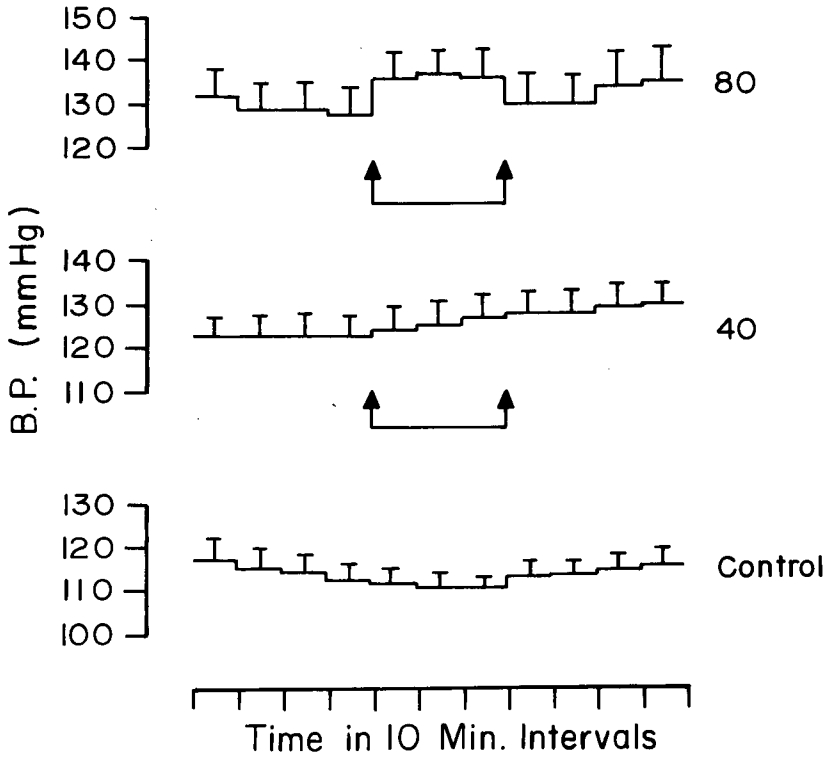

Fig. 14 Changes in systemic arterial pressure in the control and pulmonary arterial pouch distension experiments (40 and 80 cm H_2O). Average results (± SEM) taken from Fig. 11, 12 & 13.

graded distensions of the pulmonary arterial pouch elicited increases in systemic pressure proportional to the degree of distension.

The Wilcoxon two-sample rank test was used to analyse the data. For each experiment the four systemic pressure readings in period I were averaged and subtracted from the average value of the three readings in period II (II-I). The resultant values from the ten control experiments in which the pouch pressure was kept constant for the entire 110 min were compared with those where the pouch pressure was raised to 80 cm H_2O. There was a significant increase in the systemic pressure as the pouch was distended to 80 cm H_2O for 30 min when compared with the control experiments ($P<0.01$). Similarily for II-III between the same two groups of experiments $P<0.025$. Systemic blood pressure changes observed when the pulmonary arterial pouch was distended from 5-40 cm H_2O were not statistically significant.

b) Effect on urine volume

The volume of urine excreted increased with respect to time in all three sets of experiments. The average initial value was approximately 0.8 ml/min, and in the case of the control experiments it reached about 1.3 ml/min at the end of the 110 min. The rate of increase was steady over the whole period. In the experiments in which the pouch was

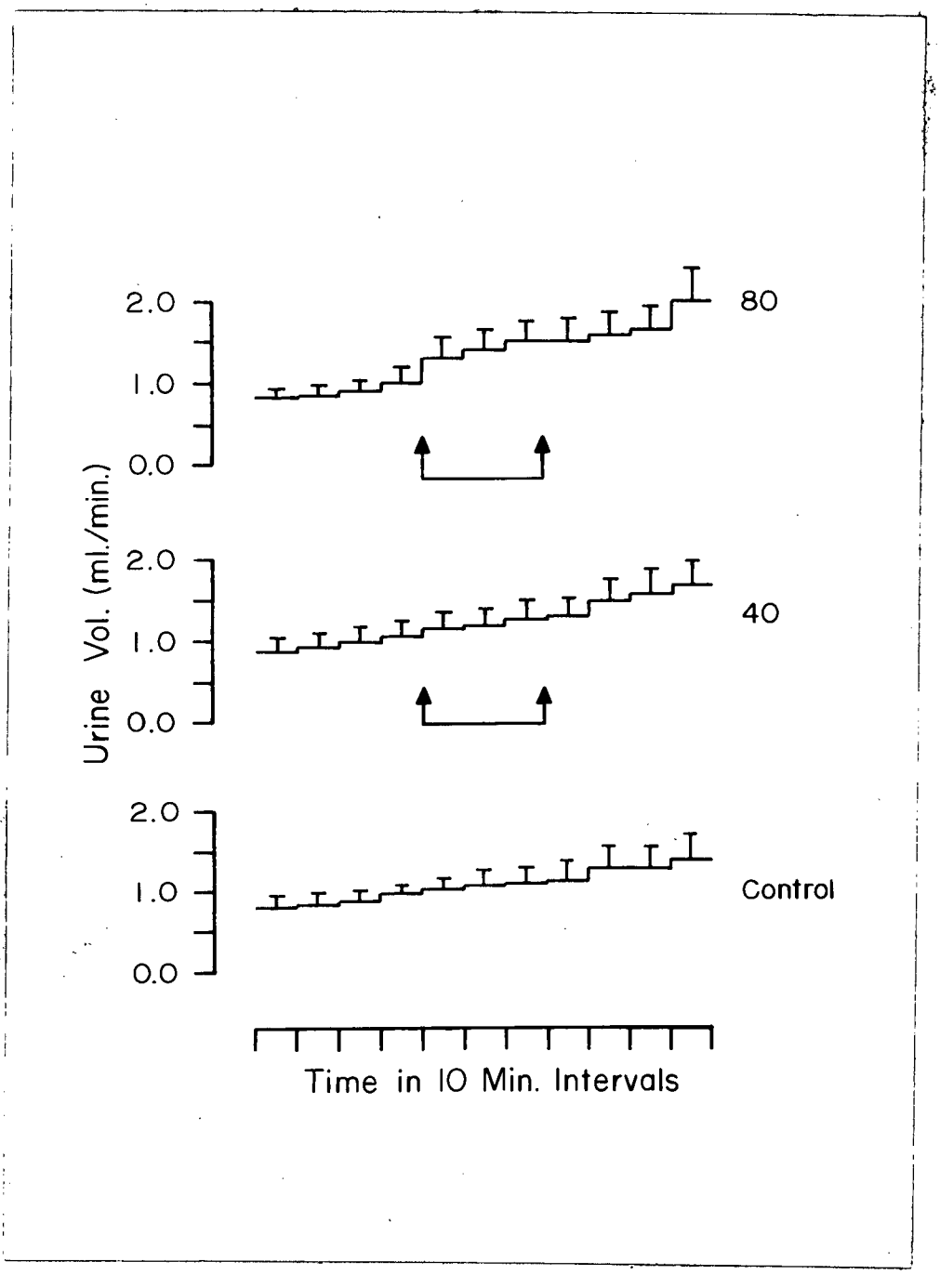

Fig. 15 Changes in urine volume of the control and pulmonary arterial pouch distension experiments (distending to 40 and 80 cm H_2O). Average results (± SEM) taken from Fig. 11, 12 & 13.

distended to 40 cm H_2O there was a higher rate of increase, and at the last collection period the average urine output was 1.6 ml/min. The rate of increase was again steady. When compared with the control experiments the difference is not significant. Distension of the pouch to 80 cm H_2O led to a step increase in urine excretion (Fig. 13) when the pouch pressure was first raised, but there was no corresponding drop when the pressure was released. The urine excretion continued to increase until the average high of 2.0 ml/min was reached at the end of the experiment. Apart from the step increase the response was similar to the other two sets of experiments. Statistical analysis indicated that the changes in urine volume in the experiments in which pulmonary arterial pouch pressure was raised to 80 cm H_2O were not significantly different from those in the control experiments.

Urine volume may be regarded as the sum of free water clearance and osmotically obligated water (osmolar clearance). These variables were examined individually to assess further any possible changes in renal function in response to pulmonary artery distension.

c) Effect on osmolar clearance

Osmolar clearance was measured in all of the urine collection experiments. For the control experiments the osmolar clearance was relatively constant throughout the

entire experiment. Mean value was about 2.1 ml/min and the average change between each collection period was always less than 0.3 ml/min (Fig. 16). Increasing the pouch pressure to 40 cm H_2O did not evoke any noticeable changes in the osmolar clearance. Statistical analysis showed that there was no significant difference from the control. Distension of the pulmonary artery pouch to 80 cm H_2O resulted in a small increase in the osmolar clearance from 2.2 ml/min to about 2.6 ml/min; however after releasing the pouch pressure at the end of the 30 min distension period the osmolar clearance did not return to the predistension level. Statistical analysis showed that the increase in osmolar clearance by pulmonary artery pouch distension to 80 cm H_2O is not significantly different from the controls.

Plasma osmolarity was also analysed in all three sets of experiments. There were no observable differences between each set, and the average value was about 290 mOs/kg. Only random fluctuations of no more than 3% were recorded. In general the plasma osmolarity can be considered constant throughout any individual experiment. Plasma sodium and potassium were also measured, again except for random fluctuations of less than 5% the average values are 145 µEq/L and 5.0 µEq/L respectively. They also can be considered to be constant throughout individual experiments.

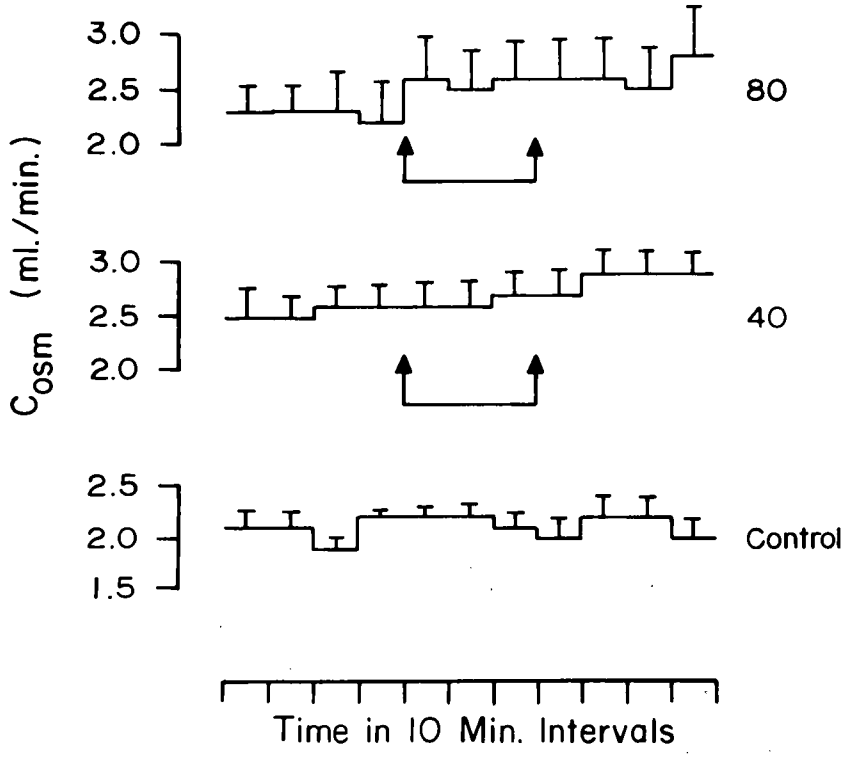

Fig. 16 Changes in urinary osmolar clearance of the control and pulmonary arterial pouch distension experiments (distending to 40 and 80 cm H_2O). Average results (± SEM) taken from Fig. 11, 12 & 13.

d) Effect on sodium excretion

The results of measurements of osmolar clearance do not indicate the content of individual solutes in the urine. Of all the solutes, sodium is of particular interest because of the importance of sodium as the major extracellular cation and its relationship to extracellular fluid volume. Changes in sodium excretion are widely accepted as possibly influencing extracellular fluid volume.

The ten control experiments showed that sodium excretion was steady around 92 µEq/min throughout the entire 110 min. No variation corresponding to time was seen. The nine experiments in which the pulmonary arterial pouch was distended to 40 cm H_2O produced a rise in sodium excretion (Fig. 17) from 127 µEq/min to 150 µEq/min and continued to rise to 168 µEq/min after the pressure was released. When compared with the control experiments, however, the change was not statistically significant. On the other hand, results from experiments where the pouch was distended to 80 cm H_2O showed almost a 50% increase in the average rate of sodium excretion from about 100 µEq/min to 148 µEq/min. This increase in period II from period I is very significant ($2P<0.02$) when compared with the control experiments. In period III of the distension experiments, the average sodium excretion rate continued to increase slightly to 160 µEq/min., when the increase in sodium excretion in period III from period I was compared between the distension and control

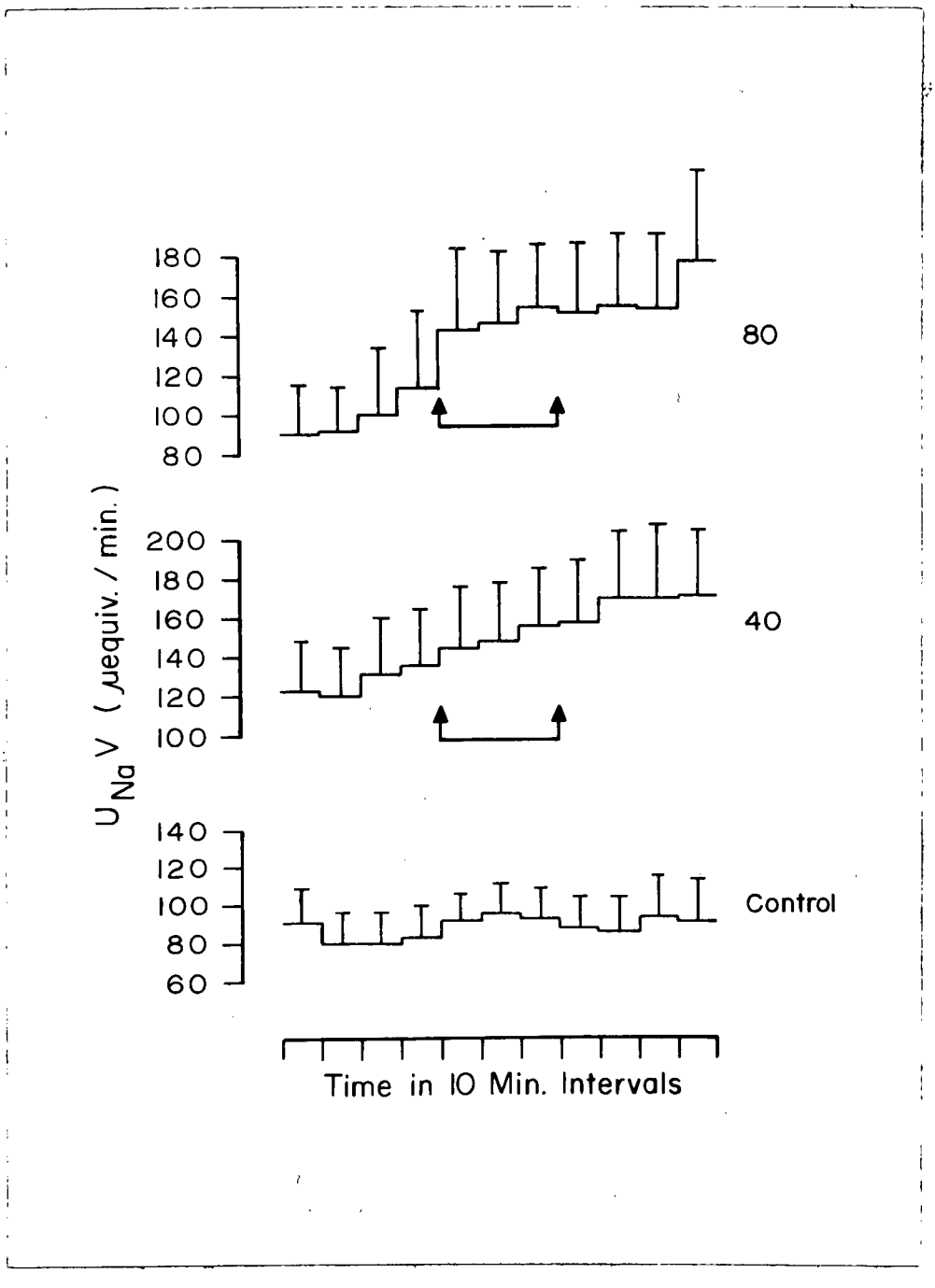

Fig. 17 Changes in urinary sodium excretion of the control and pulmonary arterial pouch distension experiments (distending to 40 and 80 cm H_2O). Average results (± SEM) taken from Fig. 11, 12 & 13.

experiments $2P>0.05$. Thus raising the pulmonary arterial pouch pressure from 5 cm H_2O to 80 cm H_2O was associated with a significant increase in sodium excretion. Although the average sodium excretion remained elevated after removal of the distension, the statistical significance of this change is doubtful.

e) Effect on potassium excretion

Potassium is the other major cation excreted in the urine besides sodium. Its regulation is closely linked to that of sodium via the aldosterone system. Along the distal tubule of the kidney aldosterone promotes the reabsorption of sodium and the excretion of potassium. In its absence, sodium excretion in the urine will increase with a corresponding decrease in the potassium excretion.

Fig. 18 shows the potassium excretion during the control experiments and the experiments with pulmonary artery distension to 40 and 80 cm H_2O. In all three cases the mean potassium excretion was approximately 140 µEq/min at the beginning of the experiments. As expected, it remained relatively constant throughout the entire 110 min experimental period, for the control experiments, with maximum variation less than 20 µEq/min. Increasing the pressure in the pulmonary artery pouch did not cause any statistically

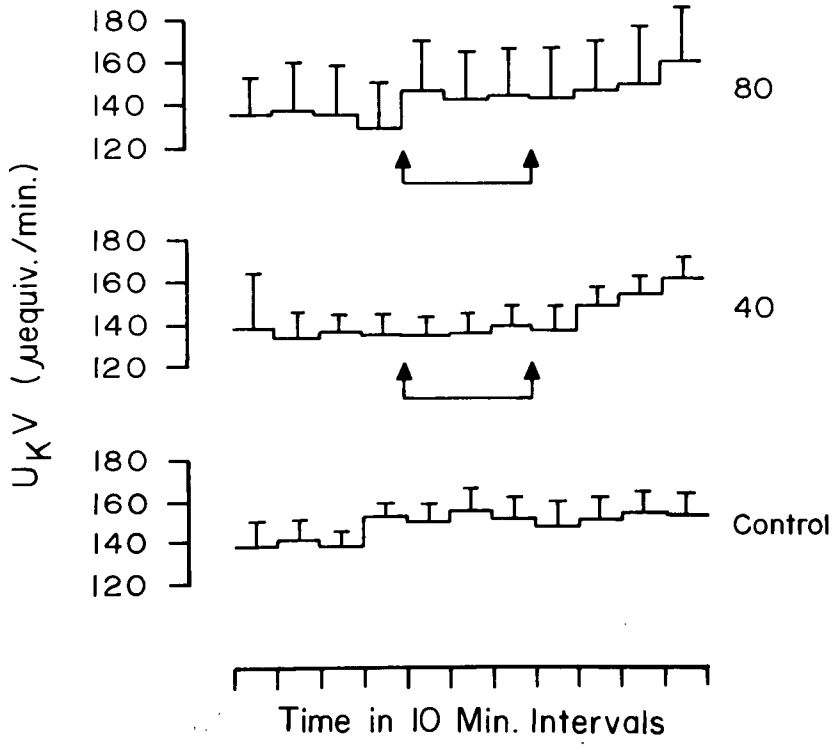

Fig. 18 Changes in urinary potassium excretion of the control and pulmonary arterial pouch distension experiments (distending to 40 and 80 cm H_2O). Average results (± SEM) taken from Fig. 11, 12 & 13.

significant changes of the mean potassium excretion. From Fig. 18 it can be seen that no single stepwise changes of more than 20 µEq/min was recorded and except for one single collection period the mean potassium excretion was always less than 16 µEq/min. Compared with the sodium excretion (Fig. 17) no correlated rise or fall of the potassium excretion was observed.

f) Effect on free water clearance

Free water clearance is a good indicator of changes in anti-diuretic hormone activity. Normally, anti-diuretic hormone promotes the reabsorption of water in the distal tubule and collecting duct to produce hypertonic urine, thus negative free water clearance.

A gradual increase in mean free water clearance was observed throughout both the control and the distension experiments (Fig. 19). The average initial value of all experiments was about -1.5 ml/min and in each series there was an increase at approximately the same rate until the average high of -0.5 ml/min was reached. There were no increases or decreases corresponding to manipulations of the pressure in the pulmonary artery pouch. As can be seen from Fig. 19 the average free water clearance of the three series of experiments was always negative, and there was no significant difference between them.

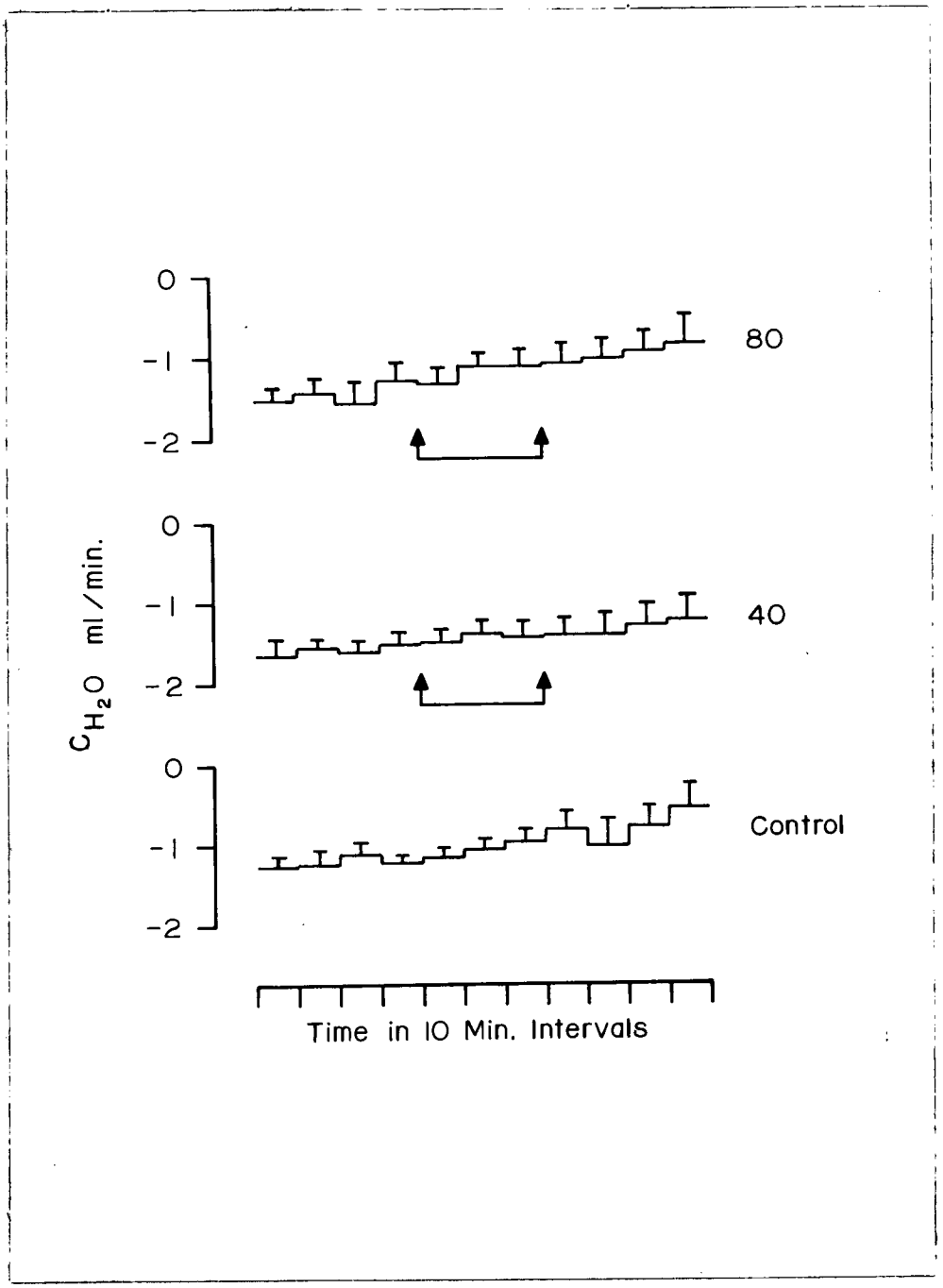

Fig. 19 Changes in urinary free water clearance of the control and pulmonary arterial pouch distension experiments (distending to 40 and 80 cm H_2O). Average results (± SEM) from same experiments shown in Fig. 11, 12 & 13.

TABLE 5

Statistical results (urine collection experiments)

	Pulmonary artery distension pressure 80 cm H_2O			Pulmonary artery distension pressure 40 cm H_2O	
Periods tested	II-I	III-I	II-III	II-I	III-I
Sys. P.	$P<0.01$	++	$P<0.025$	$P<0.05$	++
C_{os}	*	++			
C_{H_2O}	++	++		++	++
$U_{Na}V$	$2P<0.02$	*		*	*
U_KV	*	*		*	++
U_{vol}	*	*		*	++

* $2P>0.05$

++ Not tested. Believed to be not significantly different from the control by observing the raw data.

E. Urine Collection With One Kidney Isolated And Perfused

Two points emerged from the previous series of experiments. First , in order to demonstrate significant changes in urinary excretion in the relatively short time periods (for renal response), it was necessary to use high distending pressure in the pulmonary artery pouch. Secondly, the prolonged increase in sodium excretion after release of distension of the pulmonary artery pouch from 80 cm H_2O suggested that the renal response may not wholly be accounted for in terms of hemodynamic changes affecting the kidney, and suggested the possibility of a blood borne agent affecting the kidney. To test the latter possibility, six experiments were performed in which one kidney was totally isolated and perfused at constant pressure while the other kidney was intact and autoperfused. Plasma osmolarity, sodium and potassium concentrations were also measured in these six experiments. Again, except for a few random insignificant fluctuations, they can be considered constant throughout individual experiments. The average plasma osmolarity for all six experiments was 285 mOs/kg and the sodium and potassium concentrations were 145 and 5.0 μEq/L.

a) Intact kidney

Increasing the pulmonary arterial pouch pressure from 10 cm H_2O to 100 cm H_2O, then lowering it back to 10 cm H_2O, provoked a reflex increase in systemic arterial pressure.

The increase in systemic pressure from about 110 mm Hg to 125 mm Hg coincided with the rise in pulmonary arterial pouch pressure and on lowering it, the systemic pressure dropped from 116 mm Hg to 103 mm Hg. Since the cardiac output was kept constant, the changes in systemic pressure also reflected a change in systemic vascular resistance. The increase in systemic pressure is consistent with results from earlier described experiments. Paired t-test between the systemic pressure during the distension period and the average pressure of the eight collection periods before and after distension shows that the increase in systemic arterial pressure was highly significant with $P<0.005$.

Urine volume in this case showed a clear response to the pulmonary arterial pouch distension. With an initial value of 0.44 ml/min it increased to 0.66 ml/min after distension and dropped from 0.74 to 0.48 ml/min upon the release of the pressure. Paired t-test yielded P value of less than 0.0025, thus the increase in urine volume is significant.

Osmolar clearance from the intact kidney exhibited an increase in response to pulmonary arterial pressure. Before distension the average osmolar clearance of the single kidney was about 0.8 ml/min; distension to 100 cm H_2O raised the osmolar clearance to 0.96 ml/min and releasing the pressure dropped it to 0.62 ml/min. The change in osmolar clearance was synchronous with the increase in systemic arterial pressure and was statistically very

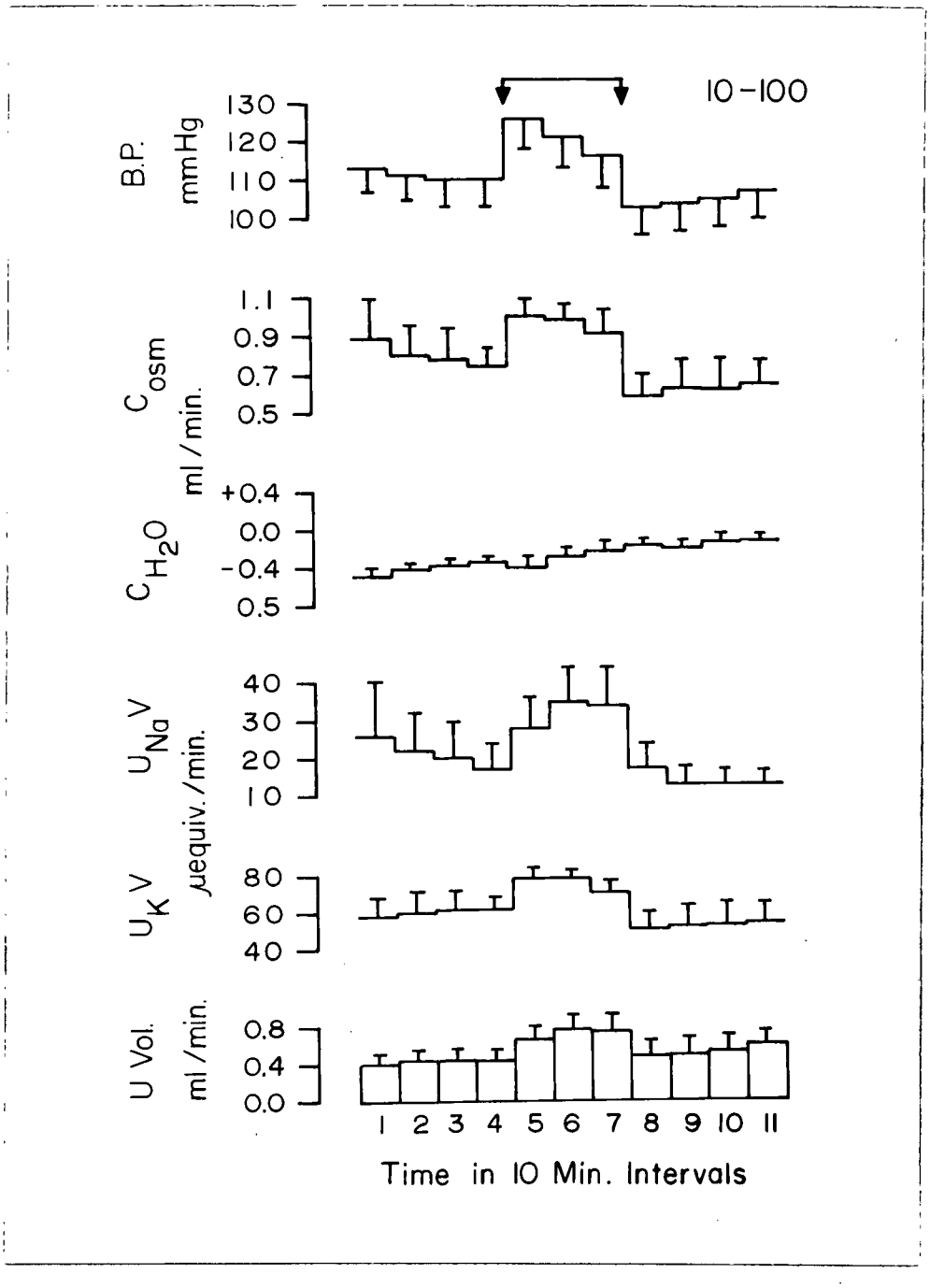

Fig. 20 Changes in systemic arterial pressure and urinary function in the intact kidney associated with pulmonary arterial pouch distension to 100 cm H_2O (periods between arrows). During the control periods before and after the distension pouch pressure was kept at 10 cm H_2O. Average results (± SEM) in 6 dogs.

TABLE 6

Kidney perfusion experiments (intact kidneys)

Time 10 min periods	Sys P mm Hg $\bar{x} \pm$ SEM		C_{os} ml/min $\bar{x} \pm$ SEM		C_{H_2O} ml/min $\bar{x} \pm$ SEM		$U_{Na}V$ µEq/min $\bar{x} \pm$ SEM		U_KV µEq/min $\bar{x} \pm$ SEM		U_{vol} ml/min $\bar{x} \pm$ SEM	
1	113	6	0.9	0.2	-0.5	0.1	26	14	58	12	0.4	0.1
2	111	6	0.8	0.2	-0.4	0.1	22	10	60	11	0.4	0.1
3	110	7	0.8	0.2	-0.3	0.1	20	10	61	11	0.4	0.1
4	110	7	0.7	0.1	-0.3	0.1	17	7	61	8	0.4	0.1
5	126	8	1.0	0.1	-0.4	0.1	28	8	78	6	0.7	0.1
6	122	8	1.0	0.1	-0.2	0.1	35	9	78	4	0.8	0.2
7	116	8	0.9	0.1	-0.2	0.1	34	10	70	7	0.7	0.2
8	103	7	0.6	0.1	-0.1	0.1	17	7	51	10	0.5	0.1
9	104	7	0.6	0.2	-0.1	0.1	13	5	52	12	0.5	0.2
10	105	7	0.6	0.2	-0.1	0.1	13	4	53	13	0.5	0.2
11	107	7	0.7	0.1	-0.1	0.1	13	4	54	11	0.6	0.2

significant (paired t-test; $P<0.0005$).

With the pouch pressure at 10 cm H_2O, sodium excretion from the intact kidney decreased steadily from 25.7 to 16.5 µEq/min. As the pressure was raised, it increased to the average of 32.0 µEq/min over the 30 minutes distension period and decreased to 13.9 µEq/min after the pouch pressure was released back to 10 cm H_2O. Paired t-test shows a significant rise in the sodium excretion ($P<0.0025$). The increase in sodium excretion in response to pulmonary pouch distension was similar to increases observed in the previous groups of experiments; unlike the previous results the output dropped about 16.5 µEq/min after releasing the pulmonary arterial pressure instead of continuing to rise.

Potassium excretion from the intact kidney responded quite differently from the experiments shown in Fig. 18. An increase of 17.3 µEq/min, from 60.9 to 78.2 µEq/min, was recorded when the pulmonary arterial pressure was raised from 10 to 100 cm H_2O (Fig. 20). When the pressure was released, potassium excretion rate decreased from 69.8 to 50.8 µEq/min, a decrease of 19 µEq/min. The increase in potassium excretion was statistically significant (paired t-test; $P<0.01$).

Free water clearance from the intact kidney was similar to the three sets of urine collection experiments described earlier. There was a steady increase from the beginning of the experiment to the end without any notice-

able breaks in the rate of increase which could be related to changes in the pulmonary arterial pouch pressure. Maximum average free water clearance was -0.45 ml/min and minimum average value was -0.07 ml/min, the difference was less than 0.4 ml/min.

b) Isolated kidney

The composition and the volume of urine samples were distinctly different from the intact kidney. Most of these differences can be attributed to the fact that all neural influences were disconnected and the increase in the systemic pressure could not have directly affected the isolated constant pressure perfused kidney. Theoretically any change in the urine composition and volume of the isolated kidney could only be brought about by circulating blood borne agents.

With the isolated kidney, the renal perfusion pressure was kept at an average of 123 mm Hg. The average renal blood flow decreased steadily from 274 ml/min down to 208 ml/min. The largest stepwise change was from 269 ml/min to 253 ml/min which occurred when the pulmonary artery pressure was raised from 10 to 100 cm H_2O, however, when the pressure was lowered the blood flow continued to decrease with a relatively steady rate without any apparent break in the trend. There was a gradual increase in the urine volume starting from 0.84 ml/min to 1.79 ml/min at

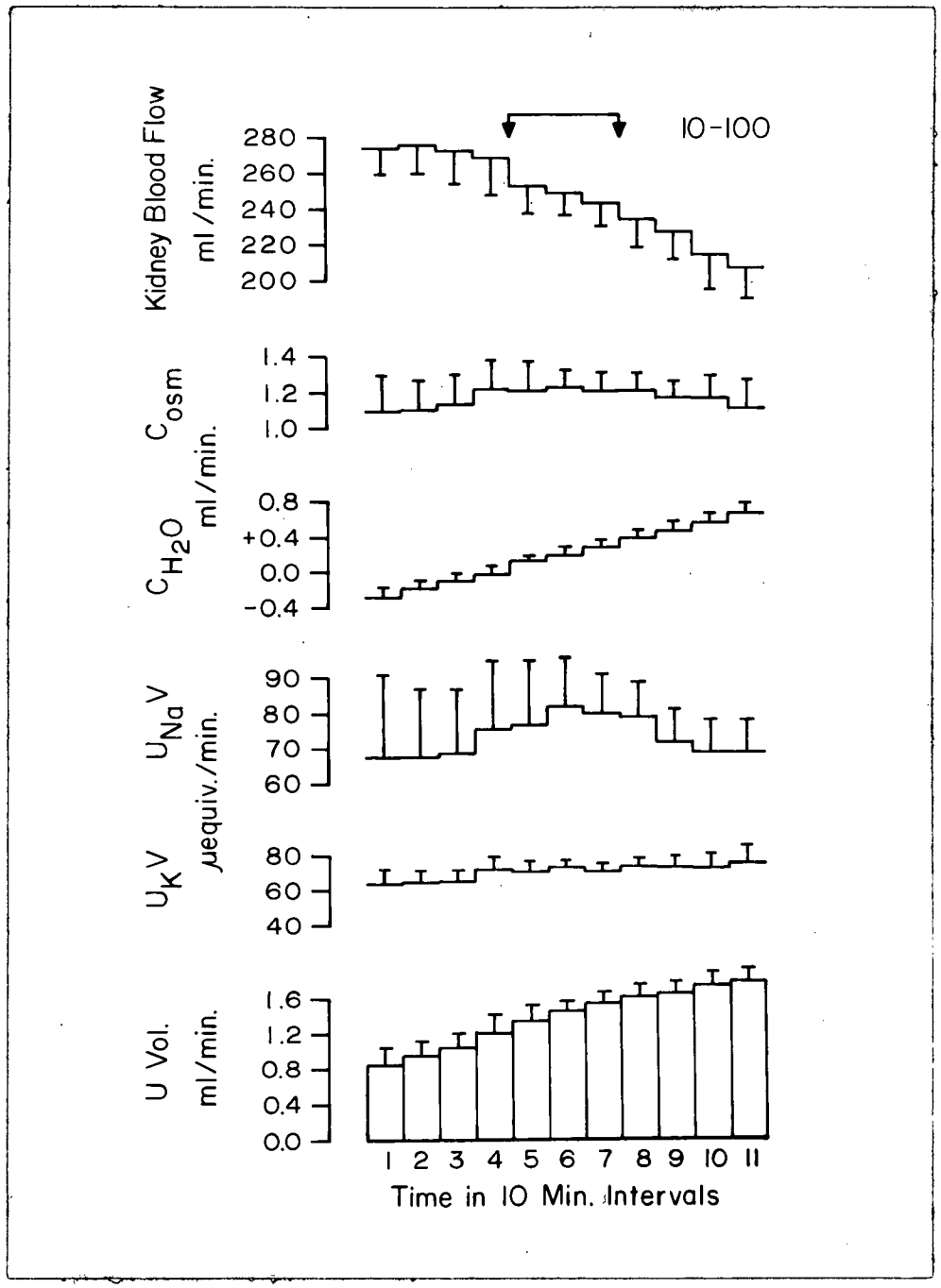

Fig. 21 Changes in renal blood flow and urinary function in the isolated constant pressure perfused (average perfusion pressure = 123 mm Hg) kidney associated with pulmonary arterial pouch distension to 100 cm H_2O. Average results (± SEM) in 6 dogs.

TABLE 7

Kidney perfusion experiments (isolated kidneys)

Time 10 min periods	RBF ml/min $\bar{x} \pm$ SEM		C_{os} ml/min $\bar{x} \pm$ SEM		C_{H_2O} ml/min $\bar{x} \pm$ SEM		$U_{Na}V$ µEq/min $\bar{x} \pm$ SEM		U_KV µEq/min $\bar{x} \pm$ SEM		U_{vol} ml/min $\bar{x} \pm$ SEM	
1	274	14	1.1	0.2	-0.3	0.1	68	23	64	8	0.8	0.2
2	276	16	1.1	0.2	-0.2	0.1	68	19	65	6	1.0	0.2
3	273	19	1.1	0.2	-0.1	0.1	69	18	65	6	1.1	0.2
4	269	21	1.2	0.2	0.0	0.1	76	19	72	7	1.2	0.2
5	253	15	1.2	0.2	0.1	0.1	77	18	71	5	1.3	0.2
6	248	12	1.2	0.1	0.2	0.1	82	14	73	4	1.5	0.1
7	243	12	1.2	0.1	0.3	0.1	80	11	71	4	1.5	0.1
8	235	16	1.2	0.1	0.4	0.1	79	10	73	5	1.6	0.1
9	228	16	1.2	0.1	0.5	0.1	73	9	73	6	1.7	0.2
10	216	19	1.2	0.1	0.6	0.1	69	9	73	8	1.7	0.2
11	209	17	1.1	0.2	0.7	0.1	69	9	75	9	1.8	0.2

RBF = Renal blood flow

Average renal perfusion pressure = 123 mm Hg

the end of the experiment. The increase was correlated to the pulmonary arterial pouch pressure.

There were small increases in sodium excretion rate during the experiment corresponding approximately to the period of pulmonary artery pouch distension. The average excretion rate before distension was 70.1 µEq/min, during the distension period it increased to 79.7 µEq/min and fell back to 72.4 µEq/min after the release of pressure. Comparison of sodium excretion rate between the distension and control periods showed no significant difference ($0.10 > P > 0.05$). These experiments do not confirm an increase in sodium excretion with pulmonary artery distension in the isolated perfused kidney.

The potassium excretion rate increased slightly over the experimental period from 63.6 µEq/min to 75.3 µEq/min. There were no changes in the excretion rate which could be related to the pulmonary artery pouch distension.

Osmolar clearance during the 110 minute experimental period was almost constant, the minimum clearance was 1.10 ml/min and the maximum clearance was 1.23 ml/min, a difference of only 0.12 ml/min. Statistical tests showed no correlation with the pulmonary arterial pouch pressure. This result is in contrast with that from the intact kidney in which the rise in pulmonary arterial pressure led to a significant increase in osmolar clearance.

Free water clearance of the isolated kidney took on the same shape as the other urine experiments, that is the clearance rose steadily throughout the experiment with no significant changes related to the pulmonary arterial pressure. However, the isolated kidney was different in one aspect, the free water clearance became positive, later in the experiment. At the beginning of the experiment the average free water clearance was -0.27 ml/min, after thirty minutes the clearance was 0.01 ml/min and at the end it was 0.68 ml/min compared with the intact kidney where the average free water clearance was always negative.

TABLE 8

Statistical results (intact and isolated kidney experiments)

	Intact Kidney		Isolated kidney
Data tested	II, $\frac{I+III}{2}$		II, $\frac{I+III}{2}$
Blood pressure	$0.0025<P<0.005$	Blood flow	*
C_{os}	$P<0.0005$		*
C_{H_2O}	$0.05<P<0.1$		*
$U_{Na}V$	$0.0005<P<0.0025$		$0.05<P<0.10$
U_KV	$0.005<P<0.01$		*
U_{vol}	$0.0005<P<0.0025$		*

* $P>0.1$

DISCUSSION

A. Review

Whereas the properties of the systemic arterial baroreceptors have been studied in considerable detail (reviewed by Heymans and Neil, 1958; Korner, 1971; Paintal, 1973; Kirchheim, 1976) there have been relatively few studies of the function of the pulmonary arterial baroreceptors. Indeed, recent reviewers (see above) have attached little importance to reflexes from this area.

a) Embryological evidence

Eberhard Koch (1931) was the first to recognize the significance of the anatomical location of the mammalian baroreceptor areas. He pointed out that these areas were developed from the embryonic visceral arch vessels and suggested that these arch vessels were all provided with corresponding visceral nerves in the embryo. The carotid sinus formed from visceral arch III is supplied by the glossopharyngeal nerve. The aortic arch baroreceptor area derived from arch IV is innervated by a branch of the superior laryngeal nerve, the nerve of visceral arch IV. He also drew attention to the vagal sensory innervation of the ductus arteriosus, a VI arch structure which develops into the main pulmonary artery and its bifurcations. From the embryological evidence the pulmonary artery baroreceptors are potentially as important as the systemic arterial receptors.

b) Histological studies

Receptors in the pulmonary artery and its main branches have been described by Dogiel (1898, 1903) and Karsner (1911) which do not have consistent distributions. Nonidez (1937, 1939) reported a pressor receptor area of the pulmonary artery bifurcation with flattened nerve fibers, ending in conspicuous rings in the pulmonary artery and which appeared to originate from the upper thoracic ganglia. Similar histological findings have also been published by Larsell and Dow (1933), Nonidez (1941), Boyd (1941), Bianconi and Green (1959) with cats, dogs, and the human beings. Nerve fibers were occasionally seen in the tunica externa of the large pulmonary arteries but never within the muscular layers. The endings are most frequently described as myelinated nerve fibers in the adventitia, terminating at the junction of the media and adventitia, or penetrating the outer layers of the media and ending as an irregular collection of coiled fibers, sometimes with swellings and enlargements on the terminal twigs. Coleridge, Kidd and Sharp (1961) reported the existence of a baroreceptor area in the left and right pulmonary arteries of dogs which is much more extensive than previously visualized. The receptors located in, or in contact with the media take the form of fine branching coiled nerve fibers compactly arranged within an area of connective tissue. They are supplied by myelinated fibers from the left and right vagus nerves. Physiological experiments by Bianconi and Green

(1959), Coleridge, Kidd and Sharp (1961) have shown that they respond to pressure applied to the pulmonary artery. Studies in man, dog and cat by Grigor'eva (1962) revealed sensory nerve elements in all three layers of the pulmonary artery. A few branchings of a myelinated nerve terminating in a large area of criss-cross unmyelinated fibers were found in the adventitia. Similar receptors were found in the media. In addition many fibers with bushy endings are in direct contact with muscular elements in all layers of the pulmonary artery.

Nonidez (1937) described an epithelioid mass in the adventitia on the dorsal aspect of the pulmonary trunk immediately caudal to its point of bifurcation. It was supplied with blood from a small branch of the aorta and a branch from the left coronary artery. Krahl (1962) reported a glomus, a homologue of the carotid and aortic glomera situated in the same position. Because of the nature of its blood supply Nonidez (1937) dismissed the idea that it might function as a chemoreceptor for venous blood. This view is supported by Verity, Hughes and Bevan (1964), who made a study of the glomus tissue in the region of the aorta and pulmonary artery in cats and found no evidence to show that the pulmonary artery supplied the tissue. They also noted that the quantity of the glomus tissue might vary from a few cells in a single lobule to large multilobular masses. Hughes (1965a, 1965b) suggested that the blood supply of the glomus tissue is derived from

the venae vasorum of the pulmonary artery and parts of the aorta; this in effect would be a portal type of circulation supplying venous blood to the glomus tissue. The significance of such a system is not known.

c) Innervation of the pulmonary artery

The nervous connections of the pulmonary artery baroreceptors were investigated by Coleridge, Kidd and Sharp (1961). Ten fibers were dissected from the right vagus, of which five were located in the recurrent cardiac nerve. This nerve is formed at about the level of the caudal cervical ganglion, receiving filaments from the ganglion, right recurrent laryngeal nerve and vagal trunk. It runs caudally and medially between the superior vena cava and trachea, and sends branches to the right pulmonary artery. The remaining right vagal fibers pass behind the superior vena cava to the right pulmonary artery in the vagal cardiac nerves which arise from the vagal trunk between the ansa subclavia and the level of the azygos vein.

A similar study was done by Bevan and Verity (1961) in cats. Using the nomenclature of Mizeres (1955) they described the recurrent cardiac nerve and sometimes the craniovagal cardiac nerves as carrying the main innervation to the pulmonary artery baroreceptors. The recurrent cardiac nerve is a moderately large nerve arising from the right recurrent laryngeal nerve as it loops around the right sub-

clavian artery. It usually receives a contribution of varying size from the vagal trunk and another from the stellate ganglion. The nerve passes dorsally to the anterior vena cava, laterally to the craniocephalic artery and arch of the aorta, to the pulmonary bifurcation where it divides into anterolateral and posterolateral branches. The anterolateral branch is the smallest. These branches fan out over the anterior and posterior aspects of the main pulmonary artery and communicate with plexi around the right and left pulmonary arteries and the pretracheal plexes.

Some pulmonary afferent fibers are the peripheral axons of cell bodies in the spinal ganglia. They reach the lungs without interruption through the stellate and middle cervical ganglia. Edgeworth (1892) traced large cardiopulmonary myelinated fibers through these sympathetic ganglia as far as the dorsal roots and suggested they might connect with the dorsal root ganglia.

Grigor'eva (1962) cut the vagi or either the nodose ganglion or the spinal ganglia C_8-T_5, as a result the baroreceptors in the adventitia and the media and the bushy nervous endings in contact with the muscular elements of the pulmonary artery degenerated. This would indicate that the sensory innervation of the area is from both the vagus nerves and the spinal nerves.

For the glomus tissue there is a rich perglomeral nerve plexus to which vagal and sympathetic fibers can be traced. Boyd (1961) described that the nervous supply is derived from the conjoined bundles of vagal and sympathetic fibers which pass towards the heart between the aorta and the pulmonary trunk. The fibers are of widely varying calibers, suggesting an extensive spectrum of fiber diameters, but whether these fibers are myelinated or not is unclear. In addition to nerve endings related to the glomus cells there is a rich nerve supply to the arterioles in the organ.

d) Electrophysiological studies

Electrophysiological studies by recording from the vagus nerves have clearly demonstrated action potentials originating from the pulmonary artery (Coleridge and Kidd, 1960). Coleridge and Kidd (1961) occluded the main pulmonary artery and observed cessation of action potential in slips of vagus nerves. When the lung roots were occluded the rate of firing of action potentials increased. Inflation of a balloon in the pulmonary artery produced similar effect. After killing the animal, they were able to trace the nerve fibers back to sites in the pulmonary artery which generated an action potential when touched. Correlating the pulmonary arterial pressure and the action potentials in the afferent vagal fibers Coleridge and Kidd (1961) observed a corresponding relationship. As the pressure

increased from an average of about 18/7 to 50/24 mm Hg (systolic/diastolic) the impulse discharge increased from a mean of approximately 1 to 26 pulses/cardiac cycle and the maximum frequency went from about 25 to 200 impulses/sec. In ten baroreceptors tested, the threshold pressures were within the range 16-25/7-13 mm Hg. They also showed that pulsatile pressure in the pulmonary artery is much more effective in producing action potentials than a steady pressure. In one of their isolated pulmonary artery preparations a steady pressure failed to generate any action potential while a pulsatile pressure with the systolic pressure less than the steady pressure did. Similar results were obtained in the intact preparations.

The large myelinated vagal fibers from the pulmonary artery actively discharge at pressures normally present in the pulmonary artery, signaling beat-to-beat changes in pressure (see above paragraph). Coleridge, Coleridge, Dangel, Kidd, Luck and Sleight (1973) measured the conduction velocity of these fibers, in five cases they range from 18.1 to 30.0 m/sec. In addition there are fine myelinated and unmyelinated C fibers arising from the pulmonary artery not connected with baroreceptors (Coleridge, Coleridge and Luke, 1965; Coleridge, Coleridge, Dangel, Kidd, Luck and Sleight, 1973). These endings had a sparse, irregular spontaneous discharge or they were quiescent. They did not respond to pressures within the physiological range but were stimulated by high pressures (60-110 mm Hg) and

the conduction velocity averaged 1.3 m/sec. These endings appear to be distributed over the main pulmonary artery and the left and right branches whereas the endings of the large myelinated fibers are located almost exclusively in the right and left main pulmonary arteries (Coleridge and Kidd, 1960; Bevan and Kinnison, 1965a, 1965b; Verity and Bevan, 1962). Coleridge et al (1973) also noticed some fibers whose endings were stimulated by both systemic hypoxia and a high pressure in their pulmonary sac (70-120 mm Hg). They suggested these were chemoreceptors (possibly the glomus cells) located on or in the pulmonary artery, the blood supply to which might be occluded by extreme distension of the pulmonary artery.

More recently Nishi, Sakanashi and Takenaka (1974), Uchida (1975) recorded action potentials from afferent fibers in the left cardiac sympathetic nerve innervating mechano-sensitive receptors in the wall of the pulmonary artery. These fibers showed irregular activity; however, a slight elevation in pulmonary arterial pressure within the range of 2.5-5/0-4 mm Hg (systolic/diastolic) elicited impulses synchronous with the systolic pressure pulse, while a further elevation of the pressure did not cause a marked increase in pulses/cardiac cycle. A sustained pressure only produced a transient burst of activity followed by a progressive decrease in the discharge. There is a possibility that these fibers represent aberrant vagal fibers, since there may be vagal contamination of the inferior cardiac

nerve. However, this is extremely unlikely because afferent fibers responding to mechanical stimulation of the pulmonary artery have also been recorded from the third ramus of the left thoracic sympathetic nerve.

e) Drug stimulation studies

The cross-sectional area of the pulmonary artery is greater than the sum of the area of the right and left branches, this phenomenon is the reverse of other major arterial bifurcations (Fry, Patel and DeFreitas, 1962). Despite the overwhelming evidence of afferent innervation of the pulmonary arteries, the reflex effects generated from these receptors are still in doubt. Porszasz, Gyorgy and Porszasz-Gibiszer (1955) observed bradycardia, hypotension and apnoea in dogs following injection of capsaicin. Bevan (1961, 1962) experimented on cats and thought that the receptors sensitive to capsaicin were located in the pulmonary artery. Analysis of possible sites of action by Porszasz, Such and Porszasz-Gibiszer (1957) found that in dogs capsaicin injections at the main pulmonary bifurcation produced the largest responses; on the other hand injection beyond the bifurcation had little or no effect. Together, with results after vagotomy, they postulated that the vagal reflex was due to activation of baroreceptors near the pulmonary bifurcation by capsaicin. However, Coleridge, Coleridge and Kidd (1964) showed capsaicin can pass freely through the pulmonary artery into the intrapulmonary vas-

cular bed despite that it was injected into the right side of the circulation only. They were able to obtain effects with injections beyond the hilum into the vascular bed of the lung itself, and sometimes the effect occurred within 1-2 sec, well within the pulmonary circulation time. Therefore they proposed that some other intrapulmonary receptors besides the pulmonary artery baroreceptors are responsive to capsaicin. This view is indirectly supported by the fact that cooling the vagus nerves to 8-10°C blocks conduction in the fibers of the extrapulmonary baroreceptors (Coleridge, Kidd and Sharp, 1961), but the effect of capsaicin was not always abolished by the same maneuver, instead it has to be cooled to 2-5°C (Coleridge, Coleridge and Kidd, 1964). The temperature differentiation suggests that afferent nervous fibers of different diameters are involved. The pulmonary artery baroreceptors with larger fibers can be blocked with a higher temperature while the other intrapulmonary receptors have smaller diameter fibers requiring a lower blocking temperature. The nature and function of these intrapulmonary receptors are not clear.

Similar cardiac and respiratory effects were reported by Bevan and Verity (1961) by using lobeline. More detailed work by Bevan (1962), Bevan and Kinnison (1965a, 1965b) revealed that small amounts (1-2 μg/kg) of lobeline injected into right side of the heart caused only ventilatory changes within 1-2 sec while larger amounts initiate in addition, bradycardia and hypotension. Atropine

reduced but not abolish the hypotension. Electrophysiological studies by Coleridge, Coleridge and Kidd (1964), Bevan and Kinnison (1965b) showed that both capsaicin and lobeline increased the afferent discharge of pulmonary artery receptors even in the absence of changes in the pulmonary artery pressure within 1-2 sec of injection into the right atrium. Verity, Hughes and Bevan (1965) explained this rapid effect by assuming that lobeline diffuses to sensory endings in the media to cause the reflex effect. Bevan speculated the ventilatory reflex response as a mechanism to lower the pulmonary arterial pressure. He stated that the immediate reflex response to a rise in pulmonary artery pressure was ventilatory inhibition and a diminution in venous return. This process would tend to lower the output of the right side of the heart and in turn the pulmonary artery pressure. The ventilatory reflex obsersed by Bevan and Kinnison (1965a) was the results of lobeline injections and there is not enough direct evidence to support this theory of pulmonary artery pressure regulation via ventilation. Moreover there are serious questions regarding how the lobeline reaches the receptors (Bevan, 1965) and whether lobeline stimulates chemoreceptors or baroreceptors.

f) Pulmonary artery distension studies

More direct stimulation of the pulmonary artery with hydraulic pressure should give a better indication of

the true physiological functions of the pulmonary artery baroreceptors. Churchill and Cope (1929) using cats, ligated the vessels of one lung and having cannulated the corresponding pulmonary artery raised the static pressure in the vessels. Bradycardia and hypotension resulted, but respiration was affected inconsistently. Experimenting with dogs, Schwiegk (1935) obtained the same results in similar experiments, furthermore he proved that the effects were reflex and were dependent on the integrity of the vagi. A fall in systemic pressure was observed when the pulmonary artery pressure was raised 10 mm Hg, the accompanied bradycardia varied from 10-24 beats per minute. Later experiments by Schweitzer (1936) and Parin (1947) were much less consistent. In Schweitzer's report, hypotension and bradycardia were observed only in two of the twelve cats tested. The static pulmonary pressures used by both Schweitzer and Parin were very high compared with the normal mean pulmonary arterial pressure. The technique used by these investigators may be critized because the rise in the pulmonary artery pressure also affected arterial, capillary and venous pressures of the pulmonary circuit. Thus any response elicited from increasing the pulmonary artery pressure can not be considered to be caused by the pulmonary artery baroreceptors alone.

Aviado, Li, Kalow, Schmidt, Turnbull, Peskin and Hess (1951) performed cross circulation experiments in which blood flowing in the right heart was isolated. Increasing

the pulmonary arterial pressure induced a vagal reflex bradycardia but no hypotension, however this claim was not supported by their experimental records. Indeed, in one of their figures which showed a rise of right heart plus pulmonary perfusion pressure there was no change in heart rate. The technique of Aviado et al (1951) allowed a more discriminative study of the pulmonary artery, however the right heart was still affected.

Lewin, Cross, Reiben and Salisbury (1961) devised a complete heart bypass with an oxygenator, and the animal was perfused with constant blood flow. The pulmonary artery was distended with blood or a balloon containing 18-28 ml of air. Inflation of the balloon led to an increase in systemic arterial pressure which was abolished by cutting or cooling (temperature unspecified) the vagus nerves. The stimuli provided by the balloon could not be quantitized and using blood the pulmonary arterial pressure had to be raised to 80-200 mm Hg to produce the response. Since the systemic blood flow was constant, the authors concluded that the response was due to vasomotor changes. Osorio and Russek (1962) introduced a balloon or a cuffed cylinder which did not impede blood flow, into the main branches of the pulmonary artery. Two types of responses were reported. With a "weak" distension they observed either no change or a fall in systemic pressure, whereas "stronger" distension caused a rise; no heart rate changes were observed. Inflation of a balloon in the pulmonary artery gives a non-specific

stimulus, for only a small portion of the balloon is actually in contact with the pulmonary arterial wall and the pressure exerted by the balloon is not measurable.

Coleridge and Kidd (1963) inserted a tube into the left main pulmonary artery of dogs and perfused the right main pulmonary artery and that portion of the left pulmonary artery outside the sleeve with controlled pressures. When the pulmonary artery sac pressure was elevated to between 20-60 mm Hg there was either no effect (10 dogs) or a fall in systemic pressure (8 dogs) and in the majority of cases (58 of 72 tests) there was no change in heart rate. When the sac pressure was raised to over 80 mm Hg the systemic pressure increased in 28 of 30 tests. An increase in respiratory movements was also noted during high pressure distension of the sac.

g) Summary

1. Embryological studies indicate the possible presence of a population of sensory endings in the pulmonary artery which is developed from the VI visceral arch.

2. Histological studies confirm the existence of receptors in the pulmonary arterial wall. They are either coiled fibers with enlargements on the terminal twigs or fibers with bushy endings. In addition glomus tissues which resemble chemoreceptors were found in the adventitia.

3. The pulmonary artery is innervated by both vagal and sympathetic afferent fibers. The receptors will degenerate if either one of these fibers is cut.

4. Electrophysiological studies have localized receptors to the right and left main pulmonary arteries. Those connected with thick myelinated fibers are active within the physiological pressure range, those connected with small myelinated or unmyelinated fibers respond only to abnormal high pressure outside the physiological range. Sympathetic activity believed to originate from the pulmonary artery has also been reported.

5. Capsaicine and lobeline stimulate receptors in the pulmonary artery and result in hypotension and ventilation inhibition. However, the location and the type of receptors (chemoreceptors/baroreceptors) stimulated is not certain.

6. Rise in the static pressure in the pulmonary artery and other vessels in the pulmonary circuit led to bradycardia and hypotension with no effect of respiration. The results are not consistent; in one report only two out of twelve animals responded.

7. Isolated right heart and pulmonary artery distension induced a vagal reflex bradycardia and no hypotension, but the evidence is not convincing.

8. "Weak" inflation of a balloon in the pulmonary artery brought a drop in the systemic pressure, a "stronger" inflation caused a rise. No heart rate responses were reported.

9. Perfusion of the main pulmonary artery, the right branch of the bifurcation and part of the left bifurcation led to a fall in systemic pressure when the perfusion pressure was between 20-60 mm Hg. With pressure above 80 mm Hg a rise in systemic pressure was observed. Again no heart rate changes were observed.

B. Cardiovascular Effect Of Pulmonary Artery Distension

It is apparent that with the exception of the experiments of Coleridge and Kidd (1963) and Lewin, Cross, Rieben and Salisbury (1961) previous studies of reflexes arising from pulmonary arterial distension have not localised the stimulus to the pulmonary artery and its right and left main branches where the receptors are known to lie. Alternatively the stimulus applied to the extra-pulmonary parts of the pulmonary artery has not been adequately controlled. The present experiments were designed to allow distension of the pulmonary artery and its right and left branches at controlled pressure whilst cardiac output was maintained constant by means of a constant flow right heart bypass. This allowed changes in systemic vascular resistance to be directly related to changes in systemic arterial pressure. Also as preliminary experiments had indicated that the system might be affected by small changes in temperature (later confirmed as described in results) the temperature of the pulmonary arterial perfusate was maintained at that of the systemic blood. It is not possible reviewing previous work to determine whether or not temperature changes in pulmonary arterial perfusion may have been a factor contributing to the variability of some of the observed effects.

There is little doubt from the results described that increasing the pressure in the pulmonary arteries

causes an increase in systemic vascular resistance. Although the changes are relatively small over the physiological range of pulmonary arterial pressure they do appear to form a continuum with the larger responses observed at higher pulmonary arterial pressures. The use of a steady pulmonary arterial pressure rather than a pulsatile one (which would have been more difficult to adequately control) is likely to underestimate the effects of small changes in pulmonary arterial pressure. For the pulmonary artery baroreceptors have been shown to be more responsive to pulsatile pressure (Coleridge and Kidd, 1961). It should also be noted that the animals had active arterial baroreceptor reflexes (as indicated by carotid occlusion) which would be likely to reduce the magnitude of the responses. The results differ markedly from those of Coleridge and Kidd (1963) in that we have shown very significant increases in systemic pressure with 40 cm H_2O (30 mm Hg) pressure in the pulmonary artery, a pressure within the range in which they observed hypotension. Our hypothesis that an increase in afferent impulse discharge from the receptors in the walls of the pulmonary arteries causes an increase in systemic vascular resistance receives support from the observation that reducing the temperature of the perfusing fluid from 37°C to 30°C was associated with a significant fall in systemic arterial pressure. This relatively small temperature change is unlikely to block nerve fibers and must be the result of a reduction in activity in receptors in or close to the pul-

monary arterial walls. Both baroreceptors and chemoreceptors have previously been shown to be affected by temperature (McQueen and Eyzaguirre, 1974; Angell James, 1971). However, it is possible that the temperature sensitive receptors are not the pulmonary arterial baroreceptors. If the effects of temperature are mediated through these baroreceptors the effects of cooling would provide evidence of a significant tonic vasoconstricter tone arising as a result of the activity of these receptors. The suggestion of Coleridge and Kidd (1963) and Coleridge et al (1973) that at high pulmonary arterial pressures there may be obstruction of blood flow to chemoreceptors lying close to the wall of the pulmonary arteries is a valid one. However, it seems unlikely that such a mechanism would account for the responses observed over the lower range of pressures used (40 cm H_2O).

The change in systemic vascular resistance observed in response to distension of the pulmonary artery has been demonstrated to be accompanied by a somewhat greater percentage increase in vascular resistance in the perfused hind-limbs but no change in the vascular resistance in the kidneys. That the sensitivity of various vascular beds to arterial baroreceptor stimulation may differ has been clearly shown (Cox and Bagshaw, 1975; Kirchheim, 1976). Renal resistance vessel changes have been described as following identical directions as muscle vessels but showing less sensitivity (Kendrick, Oberg and Wennergren, 1972a). These differences have been correlated with varying vasoconstric-

tor fiber discharge in the cat (Kendrick, Oberg and Wennergren, 1972b). Under other circumstances it has been shown that distension of the pulmonary vein-left atrial junction is associated with a decrease in renal vascular resistance (Mason and Ledsome, 1974) and a decrease in efferent sympathetic nerve activity to the kidney (Karim, Kidd, Malpus and Penna, 1972) but with no change in hind-limb vascular resistance or efferent sciatic nerve activity. The pattern of response to pulmonary arterial distension differs from both of the above in that no significant changes have been observed in renal vascular resistance in the presence of marked increases in hind-limb vascular resistance. However, since the pulmonary arteries and the left atrium both form a part of the low pressure vascular system (Gauer and Henry, 1963) the pressures in these areas are altered in a similar fashion by changes in blood volume or the distribution of the blood volume. An increase in blood volume causing a rise in pulmonary arterial and left atrial pressure could cause a rise in systemic arterial pressure with no change or a fall in renal vascular resistance. Under these circumstances an increase in sodium excretion from the kidneys would be predicted. Marked differences in renal vascular resistance have been demonstrated between dogs in which the arterial pressure has been lowered by induction of cardiogenic shock compared to those with hemorrhagic shock (Gorfinkel, Szidon, Hirsch and Fishman, 1972). In cardio-

genic shock the left atrial and pulmonary arterial pressures are raised. It is possible that reflex vascular responses arising from pulmonary arterial and left atrial receptors contribute to the differences in renal blood flow observed during cardiogenic shock compared to hemorrhagic shock.

It is apparent that neither increasing the pressure in the pulmonary arteries nor cooling the pulmonary arteries were associated with any significant changes in heart rate. The experiments may be criticized on the basis that propranolol (0.3 mg/kg) was given during the preparation. However, it was approximately 3 hrs. after the propranolol was given that the experimental records were taken and by this time complete beta-adrenergic block of the heart would not be expected. All dogs showed an increase in heart rate during carotid occlusion indicating that at least a vagal efferent pathway was intact and some dogs demonstrated sinus arrhythmia. Thus the failure to demonstrate changes in heart rate was not due to the inability of the heart to respond.

The effects of increasing the pressure in the pulmonary arteries and the effects of cooling the pulmonary arteries were wholly dependent upon the integrity of the vagus nerves. No changes in vascular resistance or heart rate were seen after cutting both vagus nerves. It is therefore likely that the afferent pathway for the reflex responses is in the vagus nerves.

Recent work using indirect techniques has suggested that impulses arising from cardiopulmonary receptors exert a tonic restraint on adrenergic discharge (Guazzi, Libretti and Zanchetti, 1962; Mancia, Donald and Shepherd, 1973; Koike, Mark, Heistad and Schmid, 1975; Mancia and Donald, 1975). The precise location of such cardiopulmonary receptors has not been identified. Our results demonstrate that receptors in or close to the walls of the pulmonary arteries generate a significant vasoconstrictor tone which may be increased by pulmonary arterial distension or decreased by cooling. In less direct experiments it may be that this vasoconstrictor effect is overwhelmed by impulses from other cardiopulmonary receptors. However, the results of experiments referred to above which compare cardiovascular reflex responses before and after cutting the vagus nerves must be interpreted with caution. It is likely that vagal section causes a much more complex series of interactions than can reasonably be described as a removal of either a net tonic inhibitory or excitatory activity.

The vasoconstrictor tone generated by distension of the pulmonary artery pouch has been shown to affect the hind-limbs but not the kidney vessels and to cause a rise in systemic pressure in the face of active arterial baroreceptor reflexes. The hypothesis that changes in blood volume or in the distribution of blood volume may affect the discharge from pulmonary artery baroreceptors and lead to modification of solute excretion by the kidney

is an attractive one and has been tested experimentally.

C. Urine Excretion From The Intact Kidneys In Response To Pulmonary Artery Distension

The kidneys respond with changes in urinary excretion only slowly to reflex changes, therefore a longer latency is usually required between the application of the stimulus and any observable changes in the urine composition. In the series of experiments described, the pulmonary artery stimulation periods were thirty minutes long hoping that it would be long enough to detect any alteration of urine excretion rate or composition. Because of the variability of urine function in individual animals comparisons of reflex responses between experimental and control animals is not always practical. A base for comparison was obtained by collecting urine samples in each animal before and after the pulmonary arterial pouch pressure was changes. Each of these control periods was forty minutes long. Since the effects of the right heart bypass and the surgical maneuvers on renal function was not known it was decided to prepare a group of animals without applying the pulmonary artery stimulus. Urine samples from this group of experiments would reflect effects of the surgical preparation. In order to make an absolute comparison with the pulmonary artery stimulation experiments these experiments are required.

Urine analysis of this type would reveal more accurate information if the experimental period could be

prolonged. Under the present conditions the length of the experiment was dictated by the volume of the blood reservoir, because hemorrhage from the surgical wounds in a heparinized animal created a problem. For a typical experiment the two liters of Dextran and Ringer Lactate mixture in the reservoir was completely lost two hours after stablization (about three and half hours after starting the right heart bypass). Increasing the volume of the artificial perfusate would allow the experiment to be prolonged, however this will further decrease the hematocrit, affect the concentrations of blood borne agents and possibly change renal function.

The urine collection experiments with both kidneys intact proved that adequate volumes of urine were produced by the preparations. Analysis of plasma samples showed that the plasma osmolarity, sodium and potassium concentrations were virtually constant throughout any single experiment and always within the normal physiological ranges. Gradual increase in free water clearance in all three sets of experiments may indicate a decrease in ADH or possibly washout of the renal medullary concentration gradient because of low hematocrit. Other than the slight change in free water clearance, systemic arterial pressure, osmolar clearance, sodium and potassium excretion were all steady throughout the control experiments. Results from these control experiments clearly demonstrated the quality of the preparation which did not seriously impair the cardiovascular

and renal functions of the animals.

Distension of the pulmonary artery pouch to 40 cm H_2O induced a small rise in the sodium excretion rate and an almost unnoticable increase in the mean systemic arterial pressure, both changes were statistically insignificant. Based on the time course of these two variables it is doubtful that the changes were results of the pulmonary pouch distensions. Osmolar clearance and potassium excretion rates were steady. Even though no significant changes in renal functions were observed in these experiments it is possible that 40 cm H_2O of steady pressure is an inadequate stimulus to initiate a renal reflex from the pulmonary artery; a higher pressure was used in another set of experiment to investigate this possibility.

Distension of the pulmonary artery pouch to 80 cm H_2O created a significant increase in sodium excretion rate. The increase in sodium excretion rate continued in some experiments even after the release of the pouch pressure. As was expected the distension also significantly raised the systemic blood pressure which returned to normal after the pouch pressure was released. Again no significant changes were observed in the osmolar and potassium excretion rates.

Examining the results from the control and the two sets of pulmonary pouch distension experiments showed that the increase in sodium excretion rate was associated with rise in systemic blood pressure (see Fig. 13). The rise

was noticeable within minutes after the pulmonary artery pouch was distended to 80 cm H_2O. The rapid nature of the increase suggested a hemodynamic effect probably secondary to cardiovascular or neural changes. The time course of the sodium excretion response is similar to O'Connor's carotid artery occlusion experiments (1955, 1958). He occluded both the carotid arteries of conscious dogs and as a result the arterial pressure increased by about 40 mm Hg accompanied by a rise in sodium excretion. O'Connor believed that the sodium response was the direct effect of the increased arterial pressure on the kidneys and differed from the effects of neurohypophysial hormones, adrenaline or adrenocortical steroids which are usually slower. The present experiments are different from the carotid artery occlusion experiments in one important aspect. The sodium excretion rate continued to increase even after the release of the pulmonary arterial pouch pressure instead of returning to the control level. The prolonged nature of the sodium response may indicate release of a circulating agent affecting sodium excretion and being released by pulmonary artery pouch distension.

Since the role of a circulating agent cannot be separately assessed in the intact kidney experiments, another series of six experiments were performed with one kidney of the animal intact and the other kidney totally isolated and perfused with controlled pressure. The isolated kidney was subjected to the influence of hormonal

factors only, for all other variables were tightly controlled. These intact and isolated kidney experiments are different from the previously discussed intact kidneys experiments in more than one way. The surgery required to prepare an isolated kidney was more extensive than that for the animals with both kidneys intact. The extra surgery created more wounded area and hemorrhage, thus a larger volume (3 liters) of artificial perfusate was needed to prime the reservoir compared with 2 liters in the intact kidneys series. The increased volume of perfusate would further change the blood composition, modifying its hemodynamic characteristics and may have a greater effect on the urine and solute excretion. Therefore no direct quantitative comparison of the two series of urine collection experiments can be made. However, between the intact and the isolated kidneys of the same animals the different influences of the hemodynamic, neural and hormal factors can be elucidated.

There is a marked difference between the results from the intact and isolated kidneys. In the intact kidney there were significant stepwise increases in the sodium, potassium excretion rates, osmolar clearance and urine volume. The increases corresponded to the pulmonary artery pouch distension and the rise in systemic arterial pressure that followed. Upon release of the pouch pressure the arterial blood pressure and the above mentioned urinary variables returned to the pre-distension level again in a stepwise manner similar to O'Connor's carotid artery

occlusion results (1958). Free water clearance is the only variable which did not seem to be affected by the pouch distension. The close resemblance with O'Connor's results confirms the significant hemodynamic effect on renal solute excretion in response to pulmonary artery distension.

The isolated kidney offered an excellent opportunity to study separately the hemodynamic and hormonal effects on the kidney. Under the influence of blood borne agents alone there were no significant changes in any of the urinary variables in response to pulmonary artery pouch distension, only the sodium excretion rate showed a small increase corresponding approximately to the distension. Since the increase is not statistically significant we can not conclude that the pouch distension releases a blood borne agent which influences the sodium excretion rate.

Renal function is affected by many different factors, generally they can be divided into three groups: hemodynamic, neural and hormonal factors. The results of the urine collection experiments will be analysed in terms of these factors.

As early as 1925, Starling and Verney recognized the relationship between renal perfusion pressure and the excretion of water and chloride. Using a heart-lung-kidney preparation they concluded that there is a direct relationship between perfusion pressure, glomerular filtration rate, water and chloride excretion. Selkurt, Hall and Spencer

(1949), Pitts and Duggan (1950) studied the effect of reduced renal perfusion pressure on sodium excretion by the in situ dog kidney and observed increases in tubular fractional sodium reabsorption as filtration rate was decreased. Thus the effect of decreased perfusion pressure in reducing sodium excretion was not attributed solely to a reduction in the filtered load of sodium. Increasing the renal perfusion pressure on the other hand depresses tubular sodium reabsorption, Selkurt (1951), Selkurt, Womack and Dailey (1965) reported an increased sodium excretion without an equivalent rise in filtration rate as perfusion pressure was heightened in the dog kidney.

In the same heart-lung-kidney preparation, Starling and Verney (1925) also observed increased sodium chloride excretion when isotonic saline was added to the blood perfusing the preparation. They referred to this effect as dilutional diuresis and attributed it to an increased glomerular filtration rate. More recently, Craig, Mills, Osbaldiston and Wise (1966), Nizet (1968), Nizet, Godon and Mahieu (1968) have observed increased sodium excretion by isolated perfused kidneys when the perfusing blood is diluted by addition of isotonic saline. Nizet, Godon and Mahieu (1968) believed that this effect is caused by a decrease in plasma protein concentration through an intrarenal mechanism. However, with the evidence avaliable we can not conclude that the increase in sodium excretion is due to the dilution of any single constituent in the blood.

Another factor that is affected by blood dilution is the hematocrit. Martino and Earley (1967) suggested that a reduction in the hematocrit would decrease the viscosity of the blood which in turn would reduce the internal resistance to flow through the renal circulation. Both Craig, Mills, Osbaldiston and Wise (1966), Nizet, Godon and Mahieu (1968) reported increased sodium excretion and solute-free water clearance in association with a fall in hematocrit when isotonic saline was added to the blood perfusing the isolated dog kidney. But in their studies the concentration of protein in plasma was decreased also, therefore the increased sodium excretion cannot be attributed entirely to either protein or red blood cell concentration. Better controlled studies by Nashat and Portal (1967), Nashat, Scholefielf, Tappin and Wilcox (1969) decreased the hematocrit in dogs without a decrease in plasma protein concentration or increase in blood volume. They observed an increased urine flow, sodium excretion and renal plasma flow. However, they did not find a decrease in sodium excretion as hematocrit was increased.

In order to perform a right heart bypass in the present experiments the extracorporeal circuit was primed with physiological fluid. As has been mentioned earlier, either two or three liters of Dextran and Ringer Lactate mixture were needed for the constant temperature reservoir in the urine collection experiments. Since the average weight of the dogs used was only about 22 kg the added

artificial perfusate should have a significant dilutional effect on the plasma protein concentration and lowered the hematocrit to below 30%. Either one or both of these mechanisms is expected to increase the sodium excretion rate and the free water clearance. Since pulmonary artery pouch distension is believed to increase the sodium excretion rate it is important to be able to distinguish between the dilution-hematocrit effect and the pouch distension effect. The control experiments where the pulmonary artery pouch was not distended served this purpose well. After stablization the animals seemed to have reached an equilibrium state for sodium excretion rate as the value became more or less constant throughout the entire 110 min urine collection period. Therefore the sodium excretion response from the distension experiments must be initiated from the pulmonary artery. The free water clearance pattern in the control experiments is most interesting, it increased steadily with respect to time. Although no direct proof is avaliable, this pattern could very well be caused by the low hematocrit in the blood and the change in the renal resistance that followed. In the pulmonary artery pouch distension experiments, free water clearance pattern were the same as the controls in each case, therefore we can conclude that the pulmonary arterial distension had no effect on free water clearance.

The kidneys are richly supplied with nerve endings. The renal nerves are described as arising, in large measure,

from the coeliac plexus (De Muylder, 1952; Mitchell, 1956; Shvalev, 1966). There are also fibers reaching the renal plexus from the thoracic and upper splanchnic nerves, the intermesenteric nerves, and the superficial hypogastic plexus. Parasympathetic fibers supply the ureter, renal pelvis and collecting tubules. McKenna and Angelakos (1968a) observed in the dog that adrenergic nerves entered the kidney with the renal artery and branched with arterial supply as it divided up to the interlobular arteries and afferent arterioles to the glomerulus. In addition to the nerve fibers lying adjacent to the smooth muscle cells of the interlobar and arcuate arteries, fibers were also found in the connective tissue surrounding these vessels. Cholinergic fibers in the kidney appear to have essentially the same distribution as the adrenergic nerves (McKenna and Angelakos, 1968b), but no cholinergic fibers are found in association with the glomeruli, efferent arterioles, veins or tubules. Because of the widespread innervation of the kidney, stripping of the tissue about the renal artery destroys only a portion of the nerves; for complete denervation, fibers must be sectioned around the renal vein, the ureter, and capsule; or the kidney must be completely isolated. Despite the detailed knowledge about renal nervous innervation little is known about its function in urine output control. Since Claude Bernard's observation that ipsilateral urine flow increased when the splanchnic nerves were sectioned on one side, little real advance has been

made. One major obstacle being that results from anesthetized and conscious animals are very different. Berne (1952) compared denervated and intact kidneys in both anesthetized and conscious dogs. In the anesthetized animals the glomerular filtration rate and sodium excretion tended to be higher in the denervated kidney, whereas in the conscious animal the glomerular filtration rate and urinary sodium excretion of the innervated and the intact kidneys were the same. Berne concluded that the higher urinary sodium excretion from the denervated kidney in the anesthetized animal was due to the vasoconstricting effect of the anesthetics on the intact kidney causing a reduction in glomerular filtration rate and thus a reduction in excretion; and that the vasoconstriction was caused by impulses carried along the renal nerves. More recently, evidence has accumulated for redistribution of renal blood flow, and glomerular filtration and the importance of these factors for regulation of salt and water balance has been recognized. Neural control is thought to have a role in the regulation of intrarenal blood flow.

Pomeranz, Birtch and Barger (1968) performed experiments in which they either stimulated the renal nerves reflexly by reducing the pressure in carotid sinuses or by stimulating the splanchnic nerve while measuring the distribution of blood flow within the kidney with ^{85}K-disappearance curve. They found that mild renal nerve stimulation was associated with a redistribution of intrarenal blood

flow even though there might be no alteration in total blood flow. The perfusion of the outer cortex fell, while that of the outer medulla rose. Tuttle and Shadler (1965) used a thermal washout technique and showed that there is a redistribution of intrarenal blood flow in animals with a low urinary excretion of sodium due to salt deprivation. When urinary excretion of sodium is high they have found an increase in cortical blood flow with a decreased blood flow to the outer medulla. Sparks, Kopald, Carriere, Chimosky and Barger (1965) reported low outer renal cortex and relatively higher outer medulla blood flow in dogs with chronic congestive heart failure which is associated with salt and water retention. Furthermore Barger (1966) indicated that intrarenal distribution of blood flow is altered in congestive heart failure by abnormal adrenergic activity. This effect can be abolished by neurotransmitter blockers. Based on this evidence Barger (1966) proposed that redistribution of intrarenal blood flow can alter urinary sodium excretion and that blood flow may be controlled by renal nervous activity. Failure to demonstrate changes in total renal vascular resistance in response to pulmonary arterial pouch distension does not therefore necessarily mean that there was no changes in renal nerve activity or that there were no changes in the distribution of blood flow in the kidney.

Besides mechanical and neural factors kidney function is strongly influenced by hormonal factors. ADH is

one of the most important hormones known to regulate urine volume. It is well known that when the plasma concentration of ADH is high a large amount of solute-free water is reabsorbed in the distal tubules and the collecting ducts of the kidney, the urine excreted will be hypertonic and the volume small. Low plasma concentration of ADH led to large volume of hypotonic urine (Handler and Orloff, 1973). One mechanism that is thought to regulate the release of ADH is left atrial pressure. This is of particular interest to us because left atrial pressure reflects the blood volume in an animal, similarly an increased blood volume should also increase the pulmonary arterial pressure. Henry, Gauer and Reeves (1956) suggested that the left atrial receptors function as volume receptors which influence urine flow. Since then many groups have reported diuresis as a result of inflating a balloon in the left atrium (Ledsome, Linden and O'Connor, 1961; Lydtin and Hamilton, 1964; Ledsome and Linden, 1968; Lawrence, Ledsome and Mason, 1973). Numerous systemic and pulmonary hemodynamic changes are induced by the balloon inflation including increases in heart rate (Carswell, Hainsworth and Ledsome, 1970; Lawrence, Ledsome and Mason, 1973), pulmonary artery pressure (Henry, Gauer and Reeves, 1956) and decreases in cardiac output (Kahl, Flint and Szidon, 1974). However, the diuresis is widely believed to be caused by a decrease in the concentration of circulating ADH. Using bioassay techniques Shu'ayb, Moran and Zimmerman (1965), Johnson, Moore and

Segar (1969), Bermman, Malvin, Jochim and Roberts (1971) submitted evidence that endogenous ADH titers decrease during left atrial distension. The results presented by these groups are far from conclusive because of the low precision of the hydrated, anesthetized rat assay and have been contested by several investigators. Other groups reasoned that if suppression of endogenous ADH mediates this response, then an exogenous infusion of ADH during the period of atrial distension should abolish the diuresis. Ledsome and Mason (1972) demonstrate that infusing 0.4-1.0 m-u/kg-min of ADH can block the increase in free water clearance induced by left atrial distension. Similar results were also reported by Kinnery and DiScala (1972). In the same report Ledsome and Mason (1972) also described an increase in osmolar clearance in response to left atrial distension, infusion of ADH had no effect on this response and no explaination has been offered for this effect. Contradicting results were presented by Kappagoda, Linden, Snow and Whitaker (1974). Using their own bioassay technique which they claim to be more sensitive, they did not find a diminution in plasma ADH titers during atrial distension. Based on these results they concluded that ADH does not mediate the diuretic response of left atrial distension. A more recent study by the same group (Kappagoda, Linden, Snow and Whitaker, 1975) provided further support for their view, acute pituitary cauterization was not found to abolish the diuretic response to left atrial distension. Some earlier investigators also

doubted the role of ADH in the left atrial response. Goetz, Hermeck, Slick and Starke (1970), Goetz, Bond, Hermeck and Trank (1970) believed that if a rise in left atrial pressure causes a diuresis by suppressing ADH release, then the antidiuretic effect associated with a fall in left atrial pressure should stimulate ADH release. Using a bioassay technique they failed to demonstrate an increase in plasma ADH concentration. Because of the relatively low precision of ADH bioassay it is difficult to evaluate some of these experimental results, the recent development of radioimmunoassay procedures promises to improve the precision, specificity, and sensitivity of the measurement of ADH in plasma (Goetz, Bond and Bloxham, 1975). De Torrente, Robertson, McDonald and Schrier (1975) employed a radioimmunoassay technique and showed that in a group of anesthetized dogs left atrial distension was connected with a diuresis which was associated with a decrease in plasma ADH concentration. A second group of acutely hypophysectomized dogs showed no significant changes. Cardiac output, renal arterial pressure, glomerular filtration rate and solute excretion were comparable in the two groups; the diuresis was believed to be due to the change in plasma ADH concentration.

The operation of the renin-angiotensin-aldosterone system in the regulation of sodium balance and extracellular fluid volume are closely related with blood pressure so that regulation of these functions can be viewed tentatively as

a single coordinated process. This is because negative or positive changes in sodium balance are ordinarily accompanied by an isomotic contraction or expansion of blood volume and of the extracellular fluid. Sodium concentration does not usually change appreciably with changes in sodium balance because of the tendency to preserve isosmolarity by the removal or addition of appropriate amounts of water to the extracellular fluid (Laragh and Sealey, 1973).

Studies of the half-life of renin have been carried out by many groups (Schaechtelin, Gegoli and Gross, 1964; Lee, 1969). Using cross-circulation technique, with nephrectomized recipient rats, observing the decay of activity after a renin-releasing stimulus suggests that renin has a half-life in the circulation of about 15-20 min. However, in a study on three nephrectomized human beings Hannon, Deruyck, Joossens and Amery (1969) found a much longer renin half-life ranging from 42 to 120 min. Assaykeen, Otsuka and Ganong (1968) reported the half-life of renin to be 79 and 45 min in two dogs. Renin has no known physiological action of its own other than angiotensin liberation. Given intravenously, it characteristically produces a rise in blood pressure after a delay of 15-20 sec. The rise in pressure is gradual, and the height and the duration of the response is proportional to the amount injected, compared with angiotensin which produces an immediate response. The pressor action of renin is now explained by the kinetics of endogenous angiotensin liberation from circulating renin substrate (Lee, 1969).

In 1954, Skeggs, Mash, Kahn and Shumway discovered two forms of angiotensin. Angiotensin I is a decapeptide produced by the action of renin on the renin sustrate, angiotensin II is an octapeptide formed after the removal of the terminal histidylleucine from angiotensin I. In the absence of plasma, antiotensin I exhibits no significant vasoconstrictor properties whereas angiotensin II is a potent pressor substance. Angiotensin II has a very short biological half-life; after intravenous injection the pressor response in an assay animal lasts for only 1-3 min. Methot, Meyer, Biron, Lorain, Lagrue and Milliez (1964), Hodge, Ng and Vane (1967), Biron, Meyer and Panisset (1968) have repeatedly shown that 70% or more of a supraphysiological infusion of angiotensin II is removed in one circulation through the liver. This rapid removal of angiotensin accounts for its short half-life. Angiotensin II is the most powerful naturally occuring vasoconstrictor substance known. When infused intravenously it is approximately 50 times more potent than norepinephrine on a molar basis (Carpenter, Davis and Ayers, 1961). It produces a rise of systolic, diastolic, and mean arterial pressure; slowing of the heart rate and reduction in cardiac output; slight increase in intrathoracic blood volume, rise in pulmonary artery and pulmonary wedge pressure; a decrease in glomerular filtration, renal plasma flow, urine flow and urinary sodium excretion. Another striking physiological action

of angiotensin II is its effect on the adrenal cortex to evoke a prompt and sustained increase in aldosterone secretion.

When aldosterone is injected into the renal artery of dogs, a latent period of some 20 to 60 min follows before any effects are seen. After this latent period, sodium excretion in the urine is reduced, whereas the excretion of potassium and hydrogen ions is increased (Barger, Berlin and Tulenko, 1958; Ganong and Mulrow, 1958). The decreased sodium excretion is not accompanied by changes in glomerular filtration rate or in the serum sodium concentration. This indicates an effect on the renal tubular epithelium. Increased excretion of potassium and hydrogen ions on the other hand is associated with a concomitant decrease in their concentration in plasma and indicates a stimulation of renal tubular secretion of these ions. Often a quantitative relationship is implied between the sodium ions reabsorbed and the potassium ions secreted in the urine in response to aldosterone, in analogy with the coupled sodium for potassium transport in red cell membranes. Evidence for such a quantitative relationship in the kidney, however, is lacking. Giebisch, Klose and Malnic (1967) examined the nature of sodium reabsorption and potassium secretion in the kidney. They suggested that active sodium reabsorption from tubular lumen produces an electrical potential gradient across the tubular epithelium with the lumen negative to peritubular surface. The electrical gradient forces potassium from body

fluids into tubular urine and constitutes the process of potassium secretion. Aldosterone stimulates distal tubular reabsorption of sodium, it will increase transepithelial electrical potentials in this portion of the nephron and therefore enhance renal secretion of potassium. Thus the renal effect of aldosterone is enhancing secretion of potassium is indirect and secondary to sodium transport which explains the lack of quantitative reciprocal relationship between sodium and potassium excretion. Tait, Tait, Little and Laumas (1961) injected radioactive aldosterone into normal subjects and measured its disappearance. They reported that the half-life of aldosterone is about 30 min in a normal subject. Ayers, Davis, Lieberman, Carpenter and Berman (1962), Davis, Olichney, Brown and Binnion (1965) subsquently showed that in experimental caval occlusion, especially hepatic venous congestion, the aldosterone half-life can be prolonged. Since the liver is the main site of biological inactivation of aldosterone (Bougas, Flood, Little, Tait, Tait and Underwood, 1964), it is natural that aldosterone half-life is also prolonged in patients with hepatic cirrhosis (Coppage, Islane, Cooner and Liddle, 1962).

There remain certain effects not explained by Giebisch, Klose and Malnic's theory. Ganong and Mulrow (1958) injected aldosterone into the aorta and renal artery of the dog, they noticed an early increase in potassium excretion which preceded the more sustained decrease in sodium excretion. More significantly, Williamson (1963)

studied the effect of actinomycin D in blocking the sodium-retaining action of aldosterone on the rat kidney, he noted that the early enhanced secretion of potassium in response to aldosterone was not prevented. Fimognari, Fanestil and Edelman (1967) repeated Williamson's experiment, they observed essentially the same responses. The mechanism of this action of aldosterone on potassium secretion is not known.

One feature of the renal tubular response to the action of aldosterone that has received much attention is the sodium escape phenomenon. It was noted that prolonged administration of aldosterone or other mineralocorticoids led to an increased extracellular volume and appropriate gain in body weight. After several days, however, the subject began to excrete sodium in his urine in amounts equaling or exceeding his daily intake despite continued administration of aldosterone or mineralcorticoid (Relman and Schwartz, 1952; August, Nelson and Thorn, 1958; Strauss and Earley, 1959). The major edematous states--congestive heart failure, nephrotic syndrome, and cirrhosis--are associated with failure of the escape phenomenon to occur (August and Nelson, 1959). In most of these sodium escape studies the glomerular filtration rates either showed no significant change or a minimal change in the opposite direction to that in sodium excretion. Therefore some investigators considered the changes in sodium excretion were due to changes in tubular reabsorption. Other researchers also considered the possibility of a natriuretic

hormone regulating the sodium balance (Smith, 1957). Since the standard deviation of individual determinations of inulin clearance is 5-10%, it can not be ruled out that the change in sodium excretion is caused by small changes in glomerular filtration rate. This problem was resolved when de Wardener, Mills, Clapham and Hayter (1961) showed that in dogs receiving large amounts of salt-retaining steroids and vasopressin an infusion of saline caused a rise in urinary sodium excretion even when glomerular filtration rate was deliberately lowered by inflating a balloon placed in the thoracic aorta.

Bahlmann, McDonald, Ventom and de Wardemer (1967) expanded the blood volume with either Hartmann solution and 2.5% albumin or cross-circulation donation. An increase in urinary sodium excretion with a rise in PAH clearance was observed in both the innervated and denervated kidneys despite the fact that perfusion pressure was lowered a few millimeters of mercury by tightening an aorta snare above the origin of both renal arteries. The sodium response was not due to a dilutional effect, it is likely a circulating substance was involved. Tobian, Coffee and McCrea (1967) connected two reservoirs between an isolated rat kidney and a rat. The isolated kidney was supplied with blood from the arterial reservoir at constant pressure while the renal venous blood drained into the venous reservoir and returned to the rat. When a mixture of two-thirds blood and one-third Ringer solution was placed into the venous reservoir without

expanding the rat's blood volume, there was no increase in sodium excretion by the isolated kidney. But when the same amount of blood was infused intravenously into the rat there was usually a large rise in sodium excretion. Even though there is strong evidence of a natriuretic agent connected with blood volume expansion there is no standardized technique to assay its activity.

Cort (1966) initiated a technique whereby the rate of urine flow rate of the anesthetized water-loaded rat was measured. Lichardus, Pliska, Uhrin and Barth (1968), Cort, Dousa, Pliska, Lichardus, Safarova, Vranesic and Rudinger (1968) injected plasma concentrates that had been taken before and after body fluid expansion into hydrated rats. The injection of control plasma concentrate did not produce any significant change in urine flow, but the injection of concentrated experimental plasma obtained after volume expansion produced a significant rise. However, the concentrated extract prepared according to Cort has an osmolarity of about 3,000 mOs/kg, it has been criticized for its osmotic activity. In fact groups repeated the experiment and obtained widely variable results. Another technique of measuring the short-circuit current across a frog skin was also explored by Cort. Plain serum or plasma concentrate were placed on the serosal side of the skin and Frog Ringer solution was placed on the other. The short-circuit current across the frog skin directly relates to sodium transport across the skin. In several reports Cort and

Lichardus (1963a, 1963b), Cort (1966), Cort, Dousa, Pliska, Lichardus, Safarova, Vranesic and Rudinger (1968) claimed that the short-circuit current rose with the addition of the control serum and fell with the experimental serum. Close examination of the three reports revealed widely different time courses in response to the experimental serum samples. There is no ready explanation to this discrepancy and therefore the results are questionable.

Recently, Sealey, Kirshman and Laragh (1969), Viskoper, Czaczkes, Schwartz and Ullman (1969) measured the sodium excretion of hydrated rats to test the natriuretic activity of extracts prepared by either gel filtration or microfiltration. Sealey et al (1969) used extracts prepared from plasma and urine from man or animals maintained on a high-salt intake after saline infusion and from patients with primary aldosteronism or essential hypertension. Viskoper et al (1969) obtained extracts of urine from patients suffering from hypertension after they have been given an intravenous infusion of hypertonic saline. Both groups reported significant rise in urinary sodium excretion rate from the assay rats. Pearce and Veress (1975) obtained plasma samples from blood volume expanded rats. Gel filtration separated the samples into four fractions and each fraction was injected into hydropenic rats for natriuretic activity. Only the large protein fraction (molecular weight exceeding 30,000) of experimental plasma produced a significantly greater natriuresis than the corresponding fraction

from either control plasma or a fractionated 5.6% albumin solution. This natriuretic activity, Pearce attributed to a hormonal factor, either a protein or a protein bound moiety, generated by vascular expansion.

It has been shown that pulmonary artery pouch distension is associated with an increased systemic arterial pressure (Fig. 5, 13, 20). For the intact kidneys the renal perfusion pressure increased the same amount as the systemic pressure therefore it is expected that the renal function will be altered. As has been discussed earlier, increased renal perfusion would cause an increased sodium excretion rate by two mechanisms: increased glomerular filtration rate as a direct result of the increased renal perfusion pressure, thus presenting more sodium to the renal tubules; depression of sodium reabsorption by the renal tubules, which is not connected with the glomerular filtration rate. Fig. 13, 20 indeed showed an increased sodium excretion rate in association with a raised systemic pressure. In the two figures the pulmonary arterial pouch pressure were elevated to 80 and 100 cm H_2O respectively, and in both cases there were significant rises in the systemic arterial pressure corresponding to the pouch distension, the renal sodium excretion rates also showed significant increases. In contrast when the pouch pressure was increased to 40 cm H_2O (Fig. 12) it did not cause a significant change in the systemic arterial pressure, consequently the change in the sodium excretion was not significant.

Although the relationship between renal perfusion pressure and sodium excretion rate is most obvious, there are other factors we must consider. In the urine collection experiments the constant temperature blood reservoir was primed with 2 or 3 liters of artificial perfusate. The osmolarity of the perfusate was about 295 mOs/kg therefore an osmotic diuresis was not expected, but the extra perfusate had a dilution effect on the plasma proteins and lowered the hematocrit. Lowering of plasma protein concentration is known to increase the sodium excretion rate. Results from the control experiments helps to determine the dilution effects on the renal function. After a sufficient stablization period the sodium excretion rate of the control experiments settled to a relatively stable level (Fig. 11, 17) and no changes were observed throughout the three collection periods. In experiments where the pulmonary artery pouch was distended the sodium excretion rate in the pre-distension control period (period I) were always steady. From this result, we conclude that the sodium excretion rate changes with pulmonary artery pouch distension to 80 and 100 cm H_2O was not caused by the blood dilution effect but by the pulmonary arterial distension.

Blood hematocrit and plasma protein concentration would also affect the free water clearance. In has been described that when plasma protein concentration and hemarocrit are lowered by diluting the blood, urine volume and free water clearance are increased. This effect is thought

to be caused by physical factor changes inside the kidney as a result of the dilution, possibly gradual washout of the concentration gradient in the renal medulla and is not connected with ADH. In all the urine collection experiments, including the result from the isolated kidneys and control experiments free water clearance rose steadily over the entire collection periods (Fig. 19, 20, 21) similar to blood dilution results obtained by Craig et al (1966), Nizet et al (1968). Since we can not detect changes in the free water clearance in any set of experiment related to pulmonary artery pouch distension we assume that they are not related. The role of ADH in this case is not clear, for no direct measurement of the hormone was made, we can only compare the known effects of ADH with our experimental result. Changes in plasma osmolarity and left atrial distension are proven mechanisms affecting the release of ADH. In the experiments, the plasma osmolarity was monitored and found to be steady and within the physiological level for each individual experiment, therefore we can exclude effect of plasma osmolarity on the release of ADH in this case. Although the left atrial pressure was not recorded in all of the experiments, in a few preliminary experiments where it was monitored we did not detect any changes. The left atrial distension experiments of Ledsome et al (1961), Lydtin et al (1964), Ledsome et al (1968), Lawrance et al (1973) showed distinctive rises in free water clearance corresponding to the

distension, results from the present experiments do not bear any resemblance to them. Radioimmunoassay experiments by de Torrente et al (1975) showed conclusively that the left atrial distension is associated with a decrease in plasma ADH concentration and a rise in the free water clearance. Based on the fact that the free water clearance pattern from the present experiments is distinctively different from that of the known effects of ADH and the pulmonary artery pouch distension did not produce any corresponding changes, we concluded that the rate of ADH release is not affected by pouch distension.

The pulmonary artery pouch distension reflex is a neural reflex as evidenced by the absence of systemic pressure changes after cutting the vagus nerves. The increase in systemic vascular resistance occurring as a result of the distension is most likely due to increased sympathetic nervous activity. The same sympathetic nervous activity acting on the kidney may cause a redistribution of intrarenal blood flow in favor of the outer medulla. Although such a redistribution will increase the renal sodium excretion, its dffect on the total renal blood flow and resistance may be unnoticeable. Indeed our results showed no change in renal resistance in response to pulmonary artery pouch distension. The isolated kidney experiments provided a better view for the effects of renal nerve activity. Compared with the intact right kidney of the same animal, the isolated kidney always showed a higher sodium excretion rate and free water

clearance similar to Berne's 1952 results. These differences between the two kidneys are most likely to be due to denervation of the isolated kidney. Although results from the intact kidney experiments did not show how much of the sodium excretion response is mediated by the neural factor there is a possibility that such a mechanism does play a role.

The sodium excretion pattern in Fig. 12, 13 revealed two distinct sections in periods II and III which may represent two different control mechanisms. When the pulmonary arterial pouch pressure was increased in period II there was a fast increase in the systemic pressure and sodium excretion rate, the response was observable within minutes and was maintained throughout the entire distension period. Because of the fast nature of the response we propose that it is the result of hemodynamic changes subsequent to the increase in systemic arterial pressure, as has been discussed. When the pulmonary arterial pressure was lowered in period III, as expected,the systemic arterial pressure also lowered back to the pre-distension level, however the sodium excretion rate remained elevated, in fact it continued to rise. If the sodium reabsorption in the renal tubules were controlled by the renal perfusion pressure alone, returning the systemic arterial pressure to the control level should also bring the sodium excretion rate back to the control level in period I. Failure to return to the control level indicated a second factor other than the renal perfusion pressure exerting an overriding

effect on the sodium excretion rate. To postulate a humoral factor whose plasma concentration is affected by pulmonary artery pouch distension, the humoral factor altering renal sodium excretion rate would explain the sodium excretion pattern. A certain amount of time is required before the circulating level of a hormone can be altered. Suppose a hormone which enhances sodium excretion is released by the pulmonary artery pouch it is logical to expect the circulating level of the hormone to remain high for a period after the pouch pressure was released. As long as the plasma concentration of the hormone is high the urinary sodium excretion rate will remain high such as the case in Fig. 12 and 13.

Aldosterone is the first candidate considered for producing the elevated sodium excretion rate. The pulmonary artery pouch distension is followed by an increase in systemic arterial pressure and renal perfusion pressure. The exact mechanism controlling the release of renin is not known. It is generally believed that a rise in the renal perfusion pressure will inhibit the release of renin into the circulation. It follows that the plasma concentration of angiotensin and aldosterone will also decrease. The relation between the systemic pressure and rate of release of renin is not clear, but a rise in the systemic arterial pressure in this case of about 10-12 mm Hg is unlikely to cause a total cessation of renin release. Also the half-life of the aldosterone already in the circulation is at

least thirty minutes long which is too long to account for the response pattern. Potassium is another indicator for changes in plasma aldosterone level. When plasma aldosterone concentration drops potassium excretion rate will decrease in association with a rise in sodium excretion rate. Although we cannot expect to see a quantitative relationship between the sodium and potassium excretion rate, we do expect to see a change in the potassium excretion rate. However, this is not the case in our results (Fig. 18). Pulmonary artery distension increases sodium excretion rate without affecting potassium excretion rate. The possibility of a "natriuretic hormone", as considered by Smith (1957), being affected was investigated further.

The isolated kidney preparation was perfused at constant pressure therefore it allowed us to observe the sole effect of the hormone without the perfusion pressure effects. Results from the right intact kidney showed an increase in osmolar clearance, sodium and potassium excretion rates during the pulmonary artery pouch distension period (Fig. 20). All three variables returned to the pre-distension level upon the release of the pouch pressure. The response pattern clearly shows the effect of renal perfusion pressure because of the close relationship with the systemic arterial pressure. The dilution effect on sodium excretion was minimized by an appropriate stablization period evidenced by the steady sodium excretion rate in period I. One major difference of the sodium excretion rate

in Fig. 20 from those in Fig. 12, 13 is the drop in period III after the release of the pouch pressure. In Fig. 12 and 13, the continuous rise of the sodium excretion rate in period III encouraged us to postulate a natriuretic hormone as part of the response, however, the same pattern is absent from the results in Fig. 20. Without an assay technique for the natriuretic hormone we can only speculate on the possible discrepancy between the different sets of experiment. The experiments shown on Fig. 12 and 13, were done with 2 liters of artifical perfusate in the reservoir while those shown on Fig. 20 were done with 3 liters. The dilution caused by the perfusate is obvious, the question is, can the extra one liter account for the different responses in period III? Not knowing the dosage response character, the question cannot be answered. The isolated kidney which is not affected by the changes in systemic pressure should give us a better picture of the action of the hormone. Fig. 21 showed a rise in the sodium excretion rate corresponding approximately to the pulmonary artery distension and no change in potassium excretion rate. The sodium response is very likely to be caused by a humoral factor, however, statistical test indicated that the confidence level for the hypothesis that there was a change in sodium excretion rate is greater than 0.90 but less than 0.95. The normal acceptable level of confidence is when it is greater than 0.95, in this case we failed to prove that there is a significant humoral factor regulating the renal sodium excretion. Since the confidence level is greater

than 0.90 it is conceivable that with a better experimental technique to minimize the blood dilution, the effect of a natriuretic hormone can be proved to be significant.

In summary, the urine collection experiments showed that pulmonary artery pouch distension affects urinary function. Sodium excretion rate is most significantly affected while potassium excretion rate is not. A major part of the response is caused by an increased renal perfusion pressure secondary to the increase in systemic arterial pressure and a hormonal factor may also be involved. Aldosterone is an unlikely candidate because of the stable potassium excretion pattern in the result and the half-life of aldosterone. A yet unknown natriuretic hormone may be involved, however, its existence was not proven. The level of ADH is probably not changed by pulmonary artery pouch distension.

D. A Possible Physiological Role Of The Reflex Response To Pulmonary Arterial Distension

The concept that the body possess a mechanism which senses the fullness of the blood stream and provokes a diuretic response through the kidney was first proposed by Peters in 1935. Experimental verifications by isosmotic expansion of blood volume were attempted by Borst (1948), Zuidema, Clarke, Reeves, Gauer and Henry (1956) and many other groups, and indeed, an increased urine flow was recorded. The term "fullness" only gives a vague description of the volume of a fluid in relation with the volume of the container. For an elastic container like the cardiovascular beds, the potential capacity is best described by a pressure-volume diagram and its "fullness" defined by the hydrostatic pressure created by the blood.

Because of the widely different mechanical properties in separate parts of the cardiovascular bed, closer examination of individual sections revealed interesting characters. Hill and Barnard (1897) stressed the importance of the different distensibilities of the arterial and venous systems. They pointed out that the relatively small volume necessary to change the arterial pressure over a wide range could easily be taken out of the capacious venous system with only negligible changes of venous pressure. To quote their words, "that the vascular system is so constructed as a discontinuous system in order that great changes of arterial pressure may be brought about by vasoconstriction

without any concomitant alteration of venous or pulmonary pressure." To further analyse the distribution of blood volume and distensibility throughout the circulation, Gauer and Henry (1963) adapted the terms low- and high-pressure systems to replace the classical anatomical distinction between the pulmonary vascular bed and the greater circulation. The low-pressure system comprises the pulmonary circulation, right heart, and capacitance vessels of the systemic circulation and the high-pressure system consists of the arterial circulation. The estimated elastic resistance of the systemic arteries is 100 to 200 times that of the rest of the circulation and they contain only 10-20 percent of the total blood volume (Gauer and Henry, 1963; Gauer, Henry and Behn, 1970). About 80 percent of the circulating blood volume is contained in the vast capacious venous-pulmonary circulation.

Because of the relatively small volume and high elastic resistance fluid dynamic equilibrium in the high-pressure system rests entirely on flow resistance and the function of the cardiac output. It is well illustrated, by stopping the heart for 3 to 4 beats, during that time, the arterial pressure will have fallen to almost 30 mm Hg, while the 3 to 4 stroke volumes which have run off into the low-pressure system will have caused an increase of pressure of no more than a few centimeters water (Gauer, 1960). Within the low-pressure system, further functional subdivision into intrathoracic and capacitance vessel compartments

can be made. Making use of spirographic, plethsmographic and X-ray studies Sjostrand (1953a, 1953b), Gauer (1955), Gauer and Zuidema (1961) showed that great quantities of blood can be interchanged freely between the two compartments. Acceleration on a large centrifuge displaces blood into the dependent region of the body. After the acceleration has ceased, blood rushes back into the pulmonary circulation within seconds (Gauer and Zuidema, 1961). The pulmonary circulation acts as an important reservoir which allows the left ventricle to adjust immediately to varying loads independent of changes of venous return. The pulmonary bed also serves as an overflow reservoir for blood.

As mentioned earlier, the fullness of the blood stream mediated renal function. Borst (1948) viewed the oliguria following hemorrhage, as well as the diuresis following blood transfusion as a volume regulatory mechanism for the maintenance of an adequate cardiac output. Although it is difficult to exclude the changes in renal function as a simple change in filtration, available evidence suggests the existence of reflex mechanisms using intravascular mechanoreceptors which are stimulated by an expansion or contraction of the blood volume to induce changes of renal function. In order to localize sensitive zones within the circulation where these mechanoreceptors may exist, a number of authors have compared the effect of various maneuvers which change the distribution of the body fluid and influence urine flow. X-ray studies in man by Fenn, Otis,

Rahn, Chadwick and Hegnauer (1947), Kilburn and Sieker (1960) indicated displacement of blood into the thorax with negative pressure breathing and a depletion of blood under positive pressure breathing. High positive pressure breathing is accompanied by antidiuresis, antinatriuresis and fall in osmolar clearance similar to that found in hemorrhage (Drury, Henry and Goodman, 1947; Murdaugh, Sieker and Manfredi, 1969). On the other hand, negative pressure breathing usually produces an increase in free water clearance and in some cases natriuresis (Sieker, Gauer and Henry, 1954; Surtshin, Hoeltzenbein and White, 1955; Boylan, and Antkowiak, 1959; Hulet and Smith, 1959; Pabst and Gauer, 1959). In these experiments the release of a hormone was suspected because of the slow onset of the diuresis with the initiation of the pressure and its persistence after termination of the maneuver (Gauer, Henry, Sidker and Wendt, 1951). Also denervation of the kidney did not abolish the effects of negative pressure breathing (Surtshin, Hoeltzenbein and White, 1955).

The intrathoracic blood volume can also be altered by water immersion. Results from several groups have demonstrated that the hydrostatic pressure gradient induced by head-out water immersion results in a significant redistribution of blood volume (Arborelius, Balldin, Lilja and Lundgren, 1972; Epstein, Duncan and Meek, 1973; Lange, Lange, Echt and Gauer, 1974). In the study by Lange, Lange, Echt and Gauer (1974) a mean increase in intrathoracic

blood volume of 700 ml and an increase in heart volume of 180 ml was recorded. Consequent to the expanded intrathoracic volume, alterations in fluid and electrolyte homeostasis were induced including a significant natriuresis, diuresis, supression of the renin-aldosterone axis and ADH release (Epstein, Duncan and Fishman, 1972; Epstein, Katsikas and Duncan, 1973; Epstein, Pins and Miller, 1975). It is believed that the "volume stimulus" increased the stimulus to receptors in the intrathoracic area and caused the renal response. By introducing catheters into the right atrium and in some cases the pulmonary artery, Arborelius, Balldin, Lilja and Lundgren (1972) recorded an increase in mean right atrial and pulmonary arterial pressure of 12 mm Hg during water immersion. The figure was obtained after correction for the increase in pleural pressure thus represents the true transmural gradient. The increased pulmonary arterial pressure was accompanied by an augmented pulse pressure and an increase in both the cardiac output and systemic arterial pressure.

Saline solution infusion is another method to increase the intrathoracic blood volume. Strauss, Davis, Rosenbaum and Rossmeisl (1951), Levinsky and Lalone (1963), reported natriuresis and water diuresis after saline infusion. The renal responses were described to be similar to those of water immersion. Comparison of the results from the two different maneuvers by Epstein, Pins, Arrington, Denunzio and Engstrom (1975) indicates that immersion

up to the neck resulted in a significant natriuresis and kaliuresis which is comparable to that induced by the acute infusion of 2 liters of saline while the subject is in the seated position. However, the mechanism that caused the renal response was not known.

The relationship between atrial distension and water diuresis is well known (Carswell, Hainsworth and Ledsome, 1970; Gauer, Henry and Behn, 1970; Goetz, Hermeck, Slick and Starke, 1970). It is thought that the dilute diuresis resulted from atrial distension is mediated through ADH (detail discussed earlier). However, the atrial reflex can only explain part of the renal responses from saline infusion and water immersion; the natriuresis in particular is still unaccounted for. Increased sodium excretion as part of the pulmonary artery distension reflex would enlighten our knowledge on the body's responses to "volume stimulus". Our results indicate an effective hemodynamic mechanism to increase excretion of sodium. In addition there is a possibility that a yet unknown natriuretic hormone may also be involved.

Ordinarily, the pulmonary arterial mean pressure in man and dog averages one-fifth or one-sixth that in the systemic circulation (Cournand, 1947; Nahas, Visscher, Mather, Haddy and Warner, 1954). However, there is no fixed relationship between the pressure in the two circuits. In man, before the onset of systole, the pulmonary arterial pressure is of the order of 7 to 12 mm Hg; during systole

it rises abruptly to 20 to 30 mm Hg; the corresponding mean pressure is of the order of 12 to 15 mm Hg (Cournand, 1947; Dexter, Dow, Haynes, Whittenberger, Ferris, Goodale and Hellems, 1950). For 95% of the population, the range of the mean pulmonary arterial pressure is from 11 to 23 mm Hg (Marshall, Helmholz and Wood, 1962). In the dog, Hamilton, Woodbury and Vogt (1939) determined that the pulmonary arterial pressures tend to be somewhat higher than in a man, a mean pressure of 20 mm Hg is not unusual. In some disease states, the pulmonary arterial pressure can become much higher. Toor, Dulfano and Yahini (1959) made a detailed study of patients with pulmonary hypertension and others with aortic stenosis. In a group of 17 patients with functional group II and early III aortic stenosis (New York Heart Association Classification) mean pulmonary pressure ranged from 32 to 75 mm Hg. One interesting result from the same study is when compared with another group of 6 patients, who also had group II and early III aortic stenosis but without pulmonary hypertension, all other hemodynamic values were nearly equal, including the sodium filtration load. Despite the lack of a significant difference in the filtered sodium load values between the two groups, the tubular rejection of sodium per minute, expressed as percent of sodium load excreted, is significantly higher in the group with pulmonary hypertension. This percentage decreases in yet a third group of 10 patients who had pulmonary hypertension but showing signs of heart failure.

Extremely high pulmonary arterial pressures are sometimes seen in some type of heart defects. One study of 10 patients with total anomalous pulmonary venous connection recorded a high pulmonary artery systolic pressure of 119 mm Hg (Marshall, Helmholz and Wood, 1962). The figures indicate that the pressure range of 20-40 cm H_2O over which significant changes in systemic pressure were observed is well within the physiological range. Even the high pressure of 100 cm H_2O (75 mm Hg) used in some of the urine experiments is not unknown under pathological conditions.

The extracellular fluid volume consists of the interstitial fluid space and the plasma volume. They are usually in an equilibrium state according to the hydrostatic and osmotic pressure between the compartments. Changes in the volume or the composition in one will affect the other, in general the interstitial fluid space is determined by the rate of capillary filtration and reabsorption (Gauer, Henry and Behn, 1970). One cation of particular importance is sodium, because of the unique renal property to handle this ion. Reinhardt and Behrenbeck (1967) investigated the effect of altered sodium intake on the extracellular fluid volume in chronic dog experiments. With high salt intake (1.7 to 14 mEq/kg per day) salt was retained and the extracellular fluid volume went up. A new equilibrium was reached when the extracellular volume reached 22 percent body weight. With a sodium-poor diet (0.5 mEq/kg per day), the extracellular fluid volume went down to 16 percent of

the body weight within 4 months. These experiments demonstrated the dependence of the extracellular fluid volume on salt metabolism.

Changes in extracellular fluid volume will eventually affect the arterial blood pressure through changes in cardiac output in relation to the general peripheral resistance. With the use of systems analysis, this mechanism is well illustrated by Guyton, Coleman, Cowley, Manning, Norman and Ferguson (1974). Short-term arterial blood pressure control is primarily a nervous function, the arterial baroreceptor buffering circuit behaves as a "proportional negative feed-back" control system with finite gain (Warner and Cox, 1962; Guyton, Coleman, Cowley, Manning, Norman and Ferguson, 1974). The control system consists of sensors made up of arterial baroreceptors which send afferent signals to the central nervous system. The signals are integrated by the central nervous system and transformed into efferent signals in the sympathetic and parasympathetic nervous systems. The nervous signals will act on the effectors and change the cardiac function or modify the peripheral resistance to counteract the disturbance. The systemic arterial pressure will be brought quickly back, close to the reference set point. Because of the nature of the proportional control system the arterial pressure will never go back to the exact reference set point unless the disturbance is removed. The long-range level of arterial blood pressure can be increased or decreased as a result of changes

in three possible factors: (a) the rate of fluid intake, (b) the ability of the kidney to excrete fluid, and (c) the rate of fluid loss by normal mechanisms (Ledingham, 1971). The kidneys act as a servocontroller of the long-range level of arterial blood pressure. They operate through the fluid balance system (Coleman and Guyton, 1969) like an "integral negative feed-back" control system with infinite gain (Guyton and Coleman, 1969; Guyton, Coleman, Cowley, Manning, Norman and Ferguson, 1974). The sensors of the system are mostly the low-pressure receptors in the cardiovascular system. Afferent signals are also sent to the central nervous system to be integrated. However, the final output from the integrator can be nervous signals or a change in the circulating level of some hormone. They act on the kidneys (the effectors) to modify the renal function. The kidney's ability to control body fluid excretion makes it possible to attain infinite gain in the control system thus enabling it to bring the arterial pressure back exactly to the reference set point. Many of the arterial blood pressure control systems known to affect arterial blood pressure acutely, such as the arterial baroreceptor control system, and the renin-angiotensin system, also have direct or indirect effect on different aspects of the fluid balance system (Conway, 1966). In some cases, the two types of systems may have the same sensors, and the classification is not a clear cut matter. It is the contributions of both the long- and short-term control systems

on fluid balance through the kidneys that arterial pressure is precisely regulated.

Consider an increased body fluid volume, such as, shortly after fluid ingestion. The expanded volume will bring the extracellular fluid volume into a new equilibrium state, with the intracellular fluid volume, according to the osmolarity of the ingested fluid. Eventually the plasma and intrathoracic volume will also be increased, and the atrial and pulmonary arterial pressures are expected to rise. Consequently, an increased cardiac output and arterial blood pressure follow. There are several fronts of defence in the body to correct the body fluid and arterial pressure back to normal. The arterial baroreceptors are the most fast acting short-term control mechanism available, it will bring the arterial pressure back very close to the reference set point within seconds via a neuro reflex to protect the vital organs. The long-term control systems all involve the kidneys. The osmolar receptors and the atrial receptors work via the regulation of ADH. When the plasma osmolarity decreases, the release of ADH is inhibited, and more water will be excreted. An increased atrial pressure excites the atrial receptors, which also inhibits the release of ADH, and water diuresis results (detail discussed earlier), to restore the body fluid volume to normal. As we have reviewed earlier, maneuvers to increase the intrathoracic blood volume such as saline infusion, water immersion or posture change from orthostasis to a supine position are connected with

water diuresis plus natriuresis. Present knowledge on ADH control can only explain the water diuresis phenomenon, we believe that results from the pulmonary artery experiments provide evidence for body fluid volume control through changes in sodium excretion.

When the pulmonary arterial pressure is elevated, as the result of an increased intrathoracic blood volume the systemic vascular resistance is expected to increase, as a result of the pulmonary arterial distension. This will act to magnify the effect of changes in blood volume on blood pressure not only by an increase in the cardiac output but also by maintaining systemic vascular resistance. The hemodynamic changes brought about by the increased arterial pressure will increase the sodium excretion rate and urine flow of the kidneys. The urine control system would function more efficiently with the increased systemic arterial pressure to bring the blood volume back to equilibrium faster. As has been shown, when individual cardiovascular beds were examined it was found that the renal resistance was not affected by pulmonary artery distension, but for the same distending pressure, the hind-limb resistance always increased more than the general peripheral resistance. This means that for the same cardiac output the resistance changes would in effect redirect part of the blood flow away from the hind-limbs in favor of the kidneys, thus further increasing the renal sodium excretion rate.

Renal effects of pulmonary artery distension have been clearly demonstrated by the urine collection experiments. There is some evidence that part of the reflex changes may be mediated by a yet unknown natriuretic hormone. Since no reflex changes in free water clearance were observed in the same experiments we must consider the idea of separate control systems for water and sodium excretion in the intra-thoracic area; atrial receptors affecting free water clearance and the pulmonary arterial baroreceptors affecting sodium excretion.

SUMMARY

1. Evidence is presented for the existence of pulmonary arterial baroreceptors.

2. A preparation is described utilizing a constant flow, right heart bypass which allowed perfusion of an isolated pouch of the pulmonary arteries at controlled pressure.

3. Distension of an isolated pulmonary arterial pouch was associated with an increase in systemic vascular resistance. A marked increase in vascular resistance was demonstrated in the perfused hind-limbs but no changes were observed in a perfused innervated kidney. The changes in vascular resistance were statistically significant at a physiological range of pulmonary arterial pressures.

4. Distension of an isolated pulmonary arterial pouch was also associated with an increase in urinary sodium excretion during pouch distension. Continuation of the increased sodium excretion after removal of the pouch distension suggested the possibility of release of a circulating "natriuretic factor".

5. Experiments using animals with one intact kidney and one kidney perfused and totally isolated, confirmed an increase in sodium excretion from the intact kidney with pulmonary arterial distension but did not provide

support for the concept of a circulating "natriuretic factor".

6. A possible physiological role for the cardiovascular and renal changes observed during pulmonary arterial distension is discussed. The location of the pulmonary arterial baroreceptors within the highly distensible "low pressure" system means that the discharge from the receptors is affected by changes in circulating blood volume and its distribution. The hypothesis that an increased discharge from pulmonary arterial baroreceptors causes circulatory readjustments which result in an increase in sodium excretion and thus promote the appropriate modification of blood volume is an attractive one. However, a physiological role for the reflex responses described can not yet be claimed on the basis of this work.

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