RENAL HEMODYNAMIC STUDIES IN ACUTE RENAL FAILURE

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This thesis is dedicated to my beloved parents, who provided the motivation and the encouragement, and to my deserving teachers, who inspired and taught me.

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Renal Hemodynamic Studies in Acute Renal Failure

by Henry Y. M. Fung

ABSTRACT

The clinical syndrome of acute renal failure generally refers to the potentially reversible acute suppression of renal function which occurs in a variety of clinical settings, particularly following hypotension, major trauma, sepsis and the ingestion of nephrotoxins. The disturbances in renal function persist long after alleviation of the precipitating events, and the restoration of flow and function to other regional vascular beds. It is now generally accepted that the severely depressed renal function is secondary to a markedly reduced glomerular filtration rate presumably due to a persistent decrease in renal cortical blood flow.

Using the glycerol-induced acute renal failure model in the rat, we have demonstrated that the severity of functional impairment can be reduced by alpha adrenergic blockade using phenoxybenzamine, prior renal denervation, and by prior renal denervation with chronic salt loading. This implies that the sympathetic nervous system plays a significant role in the development of acute renal failure.

A new experimental model for the study of renal hemodynamics in acute renal failure has been developed in the dog. This involved the prolonged infusion of noradrenaline directly into the renal artery and

resulted in the production of unilateral acute renal failure. Systemic equipressor infusions of noradrenaline given intravenously, angiotensin II or vasopressin given into the renal artery did not result in similar functional impairment. This noradrenaline-induced model was found to resemble the human lesion both histologically and in the evolution of hemodynamic and functional changes. Intrarenal blood flow distribution, as assessed by the $Xenon^{133}$ washout technique, and, renal renal arteriography, demonstrated a general reduction in cortical blood flow. Prior phenoxybenzamine blockade prevented the development of noradrenaline-induced renal ischemia and renal failure. This latter finding is consistent with the hypothesis that the initiation of acute renal failure can be purely a vascular event.

Renal vascular responses to acetylcholine and volume expansion in the dog, and to phenoxybenzamine in both the dog and in man showed similar qualitative changes in acute renal failure as in normally functioning kidneys. It is proposed that the persistent increase in renal cortical vascular resistance seen in acute renal failure may be due to a change in the physiologic character of the renal vasculature. Once established, acute renal failure is probably an intrarenal event.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

ACh	Acetylcholine
ADR	Adrenaline
ATR	Atropine
C_{Cr}	Creatinine clearance
C_{H_2O}	Free water production
C_{osm}	Osmolar clearance
Cr	Creatinine
Hct	Hemotocrit
IRBD	Intrarenal blood flow distribution
NA	Noradrenaline
NS	Not significant at 5% level
PAH	p- aminohippurate
P_{Cr}	Plasma creatinine concentration
P_K	Plasma potassium concentration
P_{Na}	Plasma sodium concentration
P_{osm}	Plasma osmolality
POB	Phenoxybenzamine
RBF	Renal blood flow
RVR	Renal vascular resistance
$\%\ TRF_{Na}$	Percent tubular rejection of filtered sodium
U_{Cr}	Urine creatinine concentration

U_K	Urine potassium concentration
U_{Na}	Urine sodium concentration
U_{osm}	Urine osmolality
$\dot{V}$	Urine flow rate

SECTION I

INTRODUCTION

A. Statement of the Problem

Acute renal failure in the human is observed in a wide variety of clinical settings, most often involving multiple contributing factors such as salt depletion, hemorrhage, hemolysis, trauma or sepsis (Merrill, 1960; 1971). Irrespective of antecedent events, a fairly uniform pattern of renal impairment is recognized. This is characterized by gross reduction in glomerular filtration to levels usually less than 5% of normal (Bull et al.1950; Sevitt, 1959; Jaeinke et al. 1967). Total renal blood flow is reduced to levels of 30 to 50% of normal (Walker et al. 1963; Brun and Munck, 1964), and renal cortical blood flow is proportionately less than other areas (Hollenberg et al. 1968a; Hollenberg et al. 1970 and Ladefoged and Winkler, 1970). These hemodynamic changes are associated with reduction or absence in all modalities of renal excretory and regulatory function. Finally, these disturbances persist long after alleviation of the precipitating events, and the restoration of flow and function to other regional vascular beds. The associated disturbances in body fluid homeostasis result in a clinical entity referred to as acute renal failure.

It is now generally accepted that the above described intrarenal events in established acute renal failure involve a persisting high resistance to blood flow (Hollenberg et al. 1968a). However, neither the mechanisms responsible for their production nor for their persistence have yet been delineated. Several authors have commented on the

frequent disparity between the marked depression of renal function and the lack of concomittant structural abnormality (Brun and Munck, 1957; Sevitt, 1959; Finckh, 1962 and Olsen and Skjoldborg, 1967) and, have suggested a significant functional element as being the basic derangement. So far, due to the complex clinical situations where acute renal failure occurs, attempts to study the early events in clinical acute renal failure have been largely unrewarding. Obviously, one wishes to avoid such a complication and there are inevitably multiple unmeasured and usually uncontrollable variables which hamper objective evaluation. In addition, attempts in the human to reverse the demonstrated changes in renal hemodynamics once established have been unsuccessful, and a cause-effect relationship between these changes and the absence of renal function remains inferential.

Previous attempts to produce an experimental animal model resembling human acute renal failure have met with limited success. Those most closely mimicking the functional and histologic changes of the human counterpart have involved small animals such as the rat. These have included renal failure induced by the parenteral administration of methemoglobin (Teschan and Lawson, 1966; and Ruiz-Guinazu et al. 1967), globin (Menefee et al. 1964 and Henry et al. 1968), free hemoglobin (Mason et al. 1963), dichromate salts (Biber et al. 1968), uranium and heavy metal salts (Flanigan and Oken, 1965; and Biber et al. 1968) and glycerol (Cameron et al. 1956; Finckh, 1959; Carroll et al.

1965; and Oken et al. 1966). Micropuncture data derived from such small animal models has provided direct confirmation that a failure of glomerular filtration is responsible for the virtual absence of renal function, and further, that the reduced filtration rate is probably not due to tubular obstruction by cast formation, or to back diffusion of filtrate (Flanigan and Oken, 1965; Oken et al. 1966; Ruiz-Guinazu et al. 1967; Thiel et al. 1967; Wilson et al. 1967, 1969; Henry et al. 1968; Biber et al. 1968; Barenberg et al. 1968; Oken et al. 1970 and Flamenbaum et al. 1971). These models have also provided data suggesting a possible role for the renin-angiotensin system in the genesis of the lesion (McDonald et al. 1969; Thiel et al. 1970; and DiBona et al. 1971). However, the procedures used to change renin activity may also produce changes in sympathetic nervous system activity and potentially intracellular-extracellular ionic distribution as well, all of which might be expected to influence vascular reactivity and resistance to flow. In addition, small animal models do not readily lend themselves to the study of ongoing hemodynamic events.

While a large animal model such as the dog would obviate some of these difficulties, previous attempts have met with limited success. Studies of total renal blood flow during hemorrhagic hypotension have shown a rise in resistance to flow which can be diminished by prior denervation or modified by adrenergic agents during hemorrhage

(Brandfonbrener and Geller, 1952; Handley and Moyer, 1954; Hollenberg, 1965; Gill and Casper, 1969; and Bell and Lister, 1970). Unfortunately, with hemorrhagic hypotension, the dog either succumbs to the shock producing procedure or survives without renal sequelae. The intravenous administration of vasoactive agents such as catecholamines, angiotensin, or hemoglobin has resulted in only transient depression of renal function (Harrison et al. 1947; Goldberg, 1962; Hatcher et al. 1962; and Ruiz-Guinazu, 1971). Renal artery clamping produces irreversible cortical necrosis (Phillips and Hamilton, 1948; and Hamilton, 1948).

In summary, available evidence suggests that the primary disturbance in acute renal failure is a gross reduction of glomerular filtration. Studies to date have suggested that this is due to a maintained increase in vascular resistance, but have so far failed to define mechanisms involved in production or persistence of these phenomenon. A major barrier to the elucidation of pathogenesis has been the lack of a suitable large animal model and techniques which would permit close study of developing intrarenal events unmodified by factors taking place elsewhere in the body as a result of the procedure used to induce the renal lesion.

B. Choice of Experimental Model

To date, the most consistently reproducible models of acute renal failure have employed dehydrated rats subjected to a variety of

noxious stimuli. Of these, the glycerol-rat model initially developed by Oken and his colleagues (1966) has been most intensively studied. In this model, the histologic and functional changes observed following the intramuscular injection of glycerol, as well as the propensity to late recovery of function, closely mirrors human acute renal failure (Thiel et al. 1966). In addition, the severity of the lesion can be titrated against the dose of glycerol. Unlike models involving the parenteral administration of globin and its derivatives, dichromate, uranium or heavy metal salts, the initial renal insult following intramuscular glycerol resembles crush injuries in man. Although this model does not lend itself to direct measurement of renal hemodynamics, it is particularly suitable for studying the effects of the various pretreatment regimes on the eventual outcome of the lesion.

The wealth of hemodynamic data available in the dog, and the size of the animal make it particularly suitable for studying the role of renal blood flow in acute renal failure. Current studies in this laboratory have involved the development of a dog model of acute renal failure, using the infusion of noradrenaline into a single renal artery. This model is reproducible, closely resembles human acute renal failure functionally and histologically and obviates the difficulties of secondary systemic effects by maintaining a normally functioning contralateral kidney. It has been used for many of the studies reported in this thesis.

Finally, extrapolations derived from hypotheses based on animal experimentation to the clinical situation has to be verified ultimately in the human. For this reason, certain aspects of renal hemodynamics in established human acute renal failure are also included in this study.

C. Purpose and Scope of Study

The purpose and scope of study can be summarized as follows:

1. To design a reproducible experimental animal model which closely mimics known changes in renal function and hemodynamics observed in human acute renal failure. Also, with this model, it should be possible to make renal hemodynamic measurements with relative ease.

2. To test the hypothesis that the initiation of acute renal failure can be a purely vascular event.

3. To test the hypotheses that continuing participation of the renin-angiotensin axis or the sympathetic nervous system, alone or in combination are involved in genesis of acute renal failure.

4. To determine the renal vascular responses in acute renal failure in the dog and to compare these responses to those seen in human acute renal failure.

SECTION II

REVIEW OF LITERATURE

The clinical syndrome of acute renal failure generally refers to the potentially reversible acute suppression of renal function which occurs in a variety of clinical settings, particularly following hypotension, major trauma, sepsis and the ingestions of nephrotoxins. The clinical and experimental features of this syndrome have been extensively reviewed (Merrill, 1960, 1971; Franklin and Merrill, 1960; Brest and Moyer, 1967; Muehrcke, 1969). Characteristically, the clinical manifestations consist of an intitial period of oliguria and diminished glomerular filtration, with secondary biochemical changes of azotemia; acid-base, electrolyte and water disturbances due to failure of excretory function (Bull et al. 1950; Kiley et al. 1960). Recovery is generally heralded by the onset of a diuretic phase associated with a progressive improvement of glomerular filtration.

Clinical interest in acute renal failure probably started during World War II. The renal histopathology and the clinical manifestations of this lesion were described independently by Bywaters and Beall (1941), Lucke (1946) and Mallory (1947). In his description of "Lower nephron nephrosis", Lucke wrote: "The essential changes are selectively restricted to the lower segments of the nephrons and comprise focal degeneration or necrosis, the presence of heme casts, secondary inflammatory reaction in the surrounding stroma and thrombosis of thin walled veins". The three mechanisms of renal injury, viz., renal

ischemia, protein tubular cast obstruction and renal cellular toxins first proposed by the pioneer workers are still the subject of discussion today and the pathogenesis of this entity remains unknown.

Models for the study of acute renal failure

The complexity of the clinical settings in which acute renal failure occurs renders critical evaluation of pathogenetic mechanisms difficult in man. Consequently, numerous experimental models of acute renal failure have been developed. The modes of induction can be classified into four categories: (1) ischemia, (2) toxins, (3) glycerol and (4) vasoactive agents.

(1) Ischemia: Ischemia-induced renal dysfunction is a less than ideal experimental model for acute renal failure. Hamilton et al. (1948), Phillips and Hamilton (1948) and Moyer et al. (1957) induced renal ischemia in the dog by renal arterial clamping. This resulted in varying degrees of azotemia roughly correlated with the duration of interruption of blood flow. Recovery of renal function did not occur in uremic dogs. Neither the clinical course nor the histopathology corresponded to the human lesion. Penner and Bernheim (1940) reported cortical necrosis in the dog following renal arterial clamping. Davies (1970) reported cortical necrosis following renal arterial clamping in the rabbit. Mueller (1967) produced uremia in rats by renal arterial occlusion for a 70 minute period. This procedure was found to be associated with

a 50% mortality within 3-4 days post-induction. Surviving animals showed elevated blood urea nitrogen and altered urinary excretion. However, ultimate return of renal function did not occur. The histopathology of the ischemic lesion was that of diffuse cellular death of cortical and medullary cells, resembling acute cortical necrosis.

Production of renal failure by hemorrhagic shock in the dog has also been relatively unsuccessful (Phillips et al. 1946; Teschan and Mason, 1961). The experimental animal generally succumbed to the shock procedure or recovered without renal functional impairment.

(2) Toxins: Toxin-induced acute renal failure has been quite successful in producing a histologic and functional lesion comparable to that seen in the human. The most commonly employed animal has been the rat where parenteral administration of the globin derivatives, such as methemoglobin (Tschan and Lawson, 1966; Ruiz-Guinazu et al. 1967), globin (Menefee et al. 1964; Henry et al. 1968), and free hemoglobin (Mason et al. 1963) have been used. Characteristically, the animals showed hemoglobinuria, initial oliguria, elevated blood urea nitrogen, histologically acute tubular necrosis and subsequent functional recovery. The parenteral administration of mercuric chloride (Flanigan and Oken, 1966; Biber et al. 1968; DiBona et al.1970) and potassium dichromate (Biber et al. 1968), have also resulted in similar functional lesions in the rat. Parenteral administration of mercuric chloride (Solomon

et al. 1971) and uranium nitrate (Flamenbaum et al. 1971) in the dog have been reported to produce renal lesions characteristic of acute renal failure.

The intravenous administration of free hemoglobin or methemoglobin to larger animals such as the dog have met with less success (Harrison et al. 1947; Conn et al. 1956; Goldberg, 1962). Depression of renal function was less predictable and generally short-lived. No chronic studies were performed to determine if complete functional recovery was possible. Ruiz-Guinazu (1971) reported the successful induction of acute renal failure in the dog by infusion of methemoglobin directly into the renal artery. Acute renal failure was prevented by systemic phenoxybenzamine pretreatment. The purposeful intravenous infusion of free hemoglobin (Miller and McDonald, 1951) and distilled water (Blackburn et al. 1954) to produce intravascular hemolysis in man has been shown to be associated with transient decreases in GFR and renal plasma flow.

(3) Glycerol: Glycerol-induced hemoglobinuric acute renal failure in the rat has been used by numerous investigators, (Cameron and Finckh, 1956; Finckh, 1959; Carroll et al. 1965; Oken et al. 1966; Thiel et al. 1966, 1967, 1970; Wilson et al. 1967, 1969; and McDonald et al. 1970). The mechanism by which parenteral glycerol causes acute renal failure is poorly understood. Glycerol itself is not a nephrotoxic agent (Finckh,

1959), and hemoglobinuria per se has been shown not to cause acute renal failure in the rat (Carroll et al. 1965; Mason et al. 1963). Intramuscular administration of glycerol causes muscle damage, presumably due to the osmotic effects of glycerol, resulting in a condition similar to severe crush injuries in the human. Cameron and Finckh (1956) have shown that the degree of renal damage was dependent on the total dose rather than the concentration of glycerol used.

Both functional and histologic changes following the administration of glycerol are remarkably similar to human acute renal failure. This model is still widely employed, particularly for the purpose of assessing the effects of a variety of pretreatment regimes.

(4) Vasoactive agents: Several vasoconstricting agents including pitressin (Byrom, 1937), adrenaline (Penner and Bernheim, 1940), serotonin (Waugh and Pearl, 1964) and angiotensin II (Byrom, 1964) have been reported to result in varying degrees of histologic renal damage when administered parenterally in the rat. The functional aspects of these lesions have not been investigated in detail.

In the dog, prolonged infusion of adrenaline directly into the left renal artery following contralateral nephrectomy has been shown by Hatcher et al. (1962), to produce renal failure histologically similar to the human lesion. A small proportion of the animals recovered from renal failure after 7 days.

Mechanisms of oliguria in acute renal failure

The mechanism whereby oliguria is produced in acute renal failure has been a controversial issue. A major reason for this uncertainty has been the lack of suitable investigative techniques. Classical clearance methods were of little use in the presence of marked oliguria. Most of the theories regarding the pathogenesis of clinical and experimental acute renal failure were originally derived from histologic data. These include: 1. obstruction by tubular casts, 2. increased intrarenal interstitial pressure due to edema with resultant cessation of filtration, and 3. increased back diffusion of glomerular filtrate due to disruption of tubular cells leading to diminished urine flow. Much of the controversy has been settled with the advent of new methodology in studying experimental acute renal failure. These consisted of the application of micropuncture techniques and blood flow measurements independent of urine flow rates.

Goldberg (1962), Mason et al. (1963), and Menefee et al. (1964) proposed that the depressed glomerular filtration and oliguria were due to tubular obstruction by casts resulting in an elevated intratubular hydrostatic pressure resulting in cessation of glomerular filtration. However, Oken et al. (1966) have found that the luminal casts observed in vivo in glycerol-induced acute renal failure in the rat frequently could be washed down the tubule with isotonic saline injected into the

tubule at pressures equal to physiologic intra-tubular hydrostatic pressures. Ruiz-Guinazu et al. (1967) reported that in methemoglobin-induced renal failure in the rat, tubular collapse was present proximal to tubular casts and that intratubular pressure proximal to the site of "obstruction" was not elevated. In addition, tubular dilatation was observed distal to cast formation. In the dog, Mailloux et al. (1967) reported the production of acute renal failure in states of renal hypoperfusion following the systemic administration of low molecular dextran. It was suggested by these authors that tubular obstruction due to precipitation of the filtered dextran molecules in states of low urine flow was responsible for the renal shut-down. Diomi et al. (1970) have been unable to confirm this finding.

Iversen and Brun (1951) noted the presence of interstitial edema in acute renal failure. Merrill (1960) proposed that an increase in intrarenal tissue pressure due to interstitial edema could result in a decrease of glomerular filtration rate. However, the presence of interstitial edema was not in universal finding. De Wardener (1960) could not demonstrate an increase in wedged renal venous pressure in experimental acute renal failure. In addition, renal decapsulation in clinical acute renal failure was not associated with dramatic return of GFR (Merrill, 1960). As well, the increase in intrarenal pressure must be quite marked in order to produce complete cessation of filtration (Collier, 1971).

Oliver et al. (1951) employed microdissection techniques in a study of human cases of acute renal failure. Glomeruli were found to be intact. There was evidence of tubular cell disruption and apparent loss of tubular basement membrane integrity in a patchy distribution throughout the nephron often in association with intraluminal cast formation. These findings were interpreted as supporting the theory that the decreased urine flow rate was due to leakage of glomerular filtrate proximal to the site of obstruction without necessitating concomittant increases in intratubular pressures. This theory received further support from the studies of Bank et al. (1967) and the micropuncture studies of Steinhausen et al. (1969) in mercury-induced acute renal failure in the rat. Considerable back-diffusion of intraluminal dye was demonstrated. Furthermore, Steinhausen and co-workers showed that the intratubular administration of inulin was absorbed into the systemic circulation and subsequently excreted by the contralateral kidney. However, DiBona et al. (1970) could not demonstrate differences between inulin and mannitol clearances in mercury poisoned rats. As well, chronic saline loading partially protected these rats from mercury chloride-induced acute renal failure despite the presence of equally severe histologic lesion. These authors concluded that the inulin "leakage" reported by Steinhausen et al. could be due to artifactual changes induced by the handling of the nephron during micropuncture.

Ruiz-Guinazu et al. (1967) have also reported prolonged intraluminal retention of Lissamine green injected by micropuncture into collapsed tubules. Thus, the theory of back-diffusion cannot be entirely responsible for the production of oliguria in experimental acute renal failure.

Not all autopsy findings in acute renal failure show obvious histologic lesions. This disparity between the presence of altered histologic appearances and the depression of renal function has led to the postulate that the primary derangement in acute renal failure could be a functional event (Brun et al. 1957; Sevitt, 1959; Finckh et al. 1962; and Olsen, 1967). The majority of micropuncture data, (Flanigan and Oken, 1965; Oken et al. 1966; Ruiz-Guinazu, 1967; Thiel et al. 1967, 1970; Wilson et al. 1967, 1969; Barenburg et al. 1968) demonstrating a primary marked reduction in GFR, likely due to increased preglomerular arteriolar resistance, tend to support this view.

Renal hemodynamic studies in acute renal failure

The development of techniques for blood flow measurements which do not depend on urine collection has permitted studies of renal blood flow in oligo-anuric situations. These include the use of inert gas clearance (Thorburn et al. 1963; Aukland, 1964), dye dilution curve analysis (Kramer et al. 1960), and the microsphere method (Katz et al. 1971). The relative merits and limitations of these different methods have been discussed (Thorburn et al. 1963; Ladefoged,1966; Ladefoged

and Pedersen, 1967; Katz et al. 1971; Grunfield et al. 1971). The isotope external counting clearance method and the dye-dilution method have been used to measure renal blood flow in clinical acute renal failure.

Walker et al. (1963) using the indocyanine green dilution method, and Brun et al. (1955), by monitoring externally the rate of disappearance of Kr^{85}, have reported reductions in renal blood flow to 30-50% of normal values in acute renal failure. Studies of intrarenal blood flow distribution, as assessed by the $Xenon^{133}$ washout technique, have revealed cortical hypoperfusion in acute renal failure of mixed etiology (Hollenberg et al. 1968, 1970; Ladefoged and Winkler, 1970), clinical and experimental renal allograft rejection (Truniger et al. 1965; Retik et al. 1967a, 1967b) and in the hepato-renal syndrome (Epstein et al. 1970; Kew et al. 1971). The concept of an intrarenal shunt proposed by Trueta et al. (1947) has been modified to a re-distribution of intrarenal blood flow. Cortical ischemia was universally present in all cases of oliguria. In these studies, the degree of cortical ischemia was found to be proportional to the degree of reduction in renal function. These findings were substantiated by arteriographic studies which demonstrated diminished filling of cortical vessels.

There is now little doubt that acute renal failure is primarily due to a sustained reduction in renal cortical blood flow. Hollenberg et al. (1968) proposed that it was unnecessary to postulate tubular dis-

ruption or obstruction as being important mechanisms for the production of oliguria. Based on the data of Gertz et al. (1966), these authors suggested that a reduction of cortical blood flow to 40% of normal was sufficient to account for cessation of glomerular filtration if the increase in renal vascular resistance were mainly pre-glomerular. However, the mediators of this increase in renal vascular resistance have not been elucidated.

SECTION III

SPECIFIC EXPERIMENTS

A. Glycerol Induced Acute Renal Failure in the Rat

1. Effect of Intramuscular Glycerol on Renal Function in the Rat

Glycerol induced homoglobinuria has been used with increasing frequency by numerous investigators as a means of causing reversible acute tubular necrosis in experimental animals. The histologic lesions produced following intramuscular injections of glycerol in rats have been well described (Thiel et al. 1966). Although the exact mechanisms by which glycerol causes acute renal failure are still poorly understood (Carroll et al. 1965), micropuncture studies have implicated that a decrease in glomerular filtration, likely due to an increase in preglomerular vascular resistance was involved in the pathogenesis (Oken et al. 1966; Ruiz-Guinazu et al. 1967 and Thiel et al. 1970). Recently, Ayer and Coworkers (1971) reported renal cortical ischemia measured by a $Xenon^{133}$ washout technique (Grandchamp et al. 1971) in rats following intramuscular glycerol administration. This further supported the thesis that a primary renovasoconstriction was responsible for the observed functional lesion.

However, glycerol itself is not toxic to tubular epithelium (Finckh 1959). Hemoglobinuria per se has been shown not to cause acute renal failure in the rat (Carroll et al. 1965; Mason et al. 1963). Sympathetic overactivity due to volume depletion and pain has been suggested as a possible mechanism responsible for the initial vasoconstriction seen in the development of acute renal failure. Using the

glycerol-rat model, an attempt was made to study the contribution of the sympathetic nervous system in the pathogenesis of acute renal failure.

a. Methods:

A modification of the protocol initially described by Oken et al. (1966) was used. Female, Long Evans rats, weighing 200-350 gms were placed in individual metabolic cages 48 hours prior to experimentation. All rats were allowed free access to tap water and standard laboratory chow pellets. Daily body weights and urine volumes were recorded.

Following 24 hours of water deprivation, 28% glycerol, diluted in 0.9% saline, at a dose of 10 ml/kg B. Wt. was injected into the adductor muscles of the hind limbs in equally divided volumes under light ether anesthesia. The animals were allowed to recover, and drinking water was allowed ad libitum. Food was withheld from the period 24 to 48 hours post-glycerol administration. During this period, urine samples, free from contamination by food particles, were collected for subsequent analysis. Blood samples were obtained by cardiac puncture under pentobarbital anesthesia at the end of the urine collection period. Animals treated by intramuscular injections of 0.9% saline were used as controls. The non-paired t-test was employed for all statistical analysis. Blood and urine specimens were

analysed for: (1) Osmolality: freezing point depression - Fiske Osmometer model Mark III;

(2) Na^+ and K^+: flame photometer using lithium as internal standard - Radiometer FLM - 2;

(3) Creatinine: Colorimetry - Picric Acid, Jaffe method.

b. Results:

It has been previously reported from this laboratory (McLean and Thomson, 1970) that glycerol given in this manner was able to produce acute hemoglobinuric renal failure with functional and histologic lesions similar to those reported by Thiel and Coworkers (1966). This was characterized by initial oliguria and subsequent diuresis. By the second day, i.e. 24 to 48 hours post-glycerol injection, considerable renal impairment is evident in the glycerol treated group compared to the saline treated group as shown in Table I. The glycerol treated animals shown a mean urine flow rate 2.8 times that of the saline treated group due to the onset of the diuretic phase. However, endogenous creatinine clearances were considerably lower in the glycerol treated group, with the correspondingly higher plasma creatinine concentrations. Osmolar clearances and urine osmolalities were both significantly lower in the glycerol treated group, and mean % TRF_{Na} (% of tubular rejection of filtered sodium) was increased, indicating gross tubular impairment.

TABLE I

Effect of 28% I.M. Glycerol on Renal Function
24 to 48 Hours Post-glycerol Administration

	Saline Treated (n=10)	Glycerol Treated (n=20)	p Value
$\dot{V}$ (ml/100 gm/24 hrs.)	3.5 ± 0.5	9.7 ± 1.3	<0.01
C_{Cr} (ml/100 gm/24 hrs.)	500 ± 52	60.9 ± 21.6	<0.001
C_{osm} (ml/100 gm/24 hrs.)	12.2 ± 0.7	9.4 ± 1.3	<0.01
C_{H_2O} (ml/100 gm/24 hrs.)	-9.4 ± 0.8	13.8 ± 2.4	<0.001
U_{osm} (mOsm/kg)	1410 ± 180	426 ± 22.5	<0.001
% TRF_{Na} (%)	0.48 ± 0.07	39.48 ± 11.49	<0.001
P_{Cr} (mg%)	0.67 ± 0.05	5.65 ± 0.65	<0.001

Values are expressed as mean ± S.E.

Dose of 28% Glycerol = 10 ml/kg B.Wt.

c. Comments:

The observed functional changes following intramuscular glycerol injection were consistent and reproducible. Light microscopy revealed typical features of acute tubular necrosis (McLean and Thomson, 1970). Renal function 24 to 48 hours post-glycerol injection was used as control for subsequent experiments.

2. Effect of Prior Unilateral Renal Denervation on Glycerol Induced Acute Renal Failure in the Rat

It has been well established by conventional and histochemical fluorescence methods that the preglomerular afferent arterioles are richly innervated by sympathetic adrenergic nerve fibers (McKenna and Angelakos, 1968; Gosling, 1969; Almgard et al. 1971). Also, either direct or reflex stimulation of renal nerves, has been shown to produce a decrease in renal blood flow (Houck, 1951 and Block et al. 1952b) with a pattern of intrarenal distribution qualitatively similar to that seen in acute oliguric renal failure (Promeranz et al. 1968). Although prolonged sympathetic stimulation alone was unable to produce acute renal failure, it is tempting to suggest that sympathetic overactivity that occurs following glycerol administration may be partially responsible for the reduction of renal blood flow during the development of glycerol induced acute renal failure.

In an attempt to determine the significance of the sympathetic nervous system in the pathogenesis of glycerol induced acute renal

failure, the effects of prior renal denervation were studied.

a. Methods:

The glycerol model previously described was employed with the following modification. Under light ether anesthesia, the left kidney was denervated by mechanical striping of the renal pedicle through a left flank incision under a dissecting microscope. Rats were allowed to recover for a minimum of one week after surgery before being subjected to experimentation. Animals were sacrificed 48 hours post-glycerol injection and their renal function compared to a control group of rats which received only the standard dose of 28% glycerol.

In addition, the renal function 48 hours post-glycerol injection of 5 rats subjected to prior unilateral renal denervation and contralateral nephrectomy were compared to 4 rats subjected to a sham-denervation procedure with contralateral nephrectomy.

b. Results:

Renal function 24 to 48 hours post-glycerol in the unilaterally denervated group and the innervated controls are shown in Table II. Body weights after dehydration in the 2 groups were not significantly different. However, mean creatinine clearance and osmolar clearance were significantly higher in the denervated group, with correspondingly lower plasma creatinine concentration, and % TRF_{Na}.

TABLE II

Effect of Prior Unilateral Renal Denervation on Glycerol-induced Acute Renal Failure. Renal Function 24 to 48 Hours Post-glycerol Administration

	Control (n=20)	Prior Denervation (n=8)	p Value
$\dot{V}$ (ml/100 gm/24 hrs.)	9.7 ± 1.3	13.7 ± 2.0	NS
C_{Cr} (ml/100 gm/24 hrs.)	60.9 ± 21.6	150.4 ± 22.3	<0.05
C_{osm} (ml/100 gm/24 hrs.)	9.4 ± 1.3	13.1 ± 1.0	<0.05
C_{H_2O} (ml/100 gm/24 hrs.)	13.8 ± 2.4	18.5 ± 4.9	NS
U_{osm} (mOsm/kg)	426.1 ± 22.5	372.6 ± 74.5	NS
% TRF_{Na} (%)	39.48 ± 11.49	5.83 ± 1.54	<0.05
P_{Cr} (mg%)	5.65 ± 0.65	2.67 ± 0.45	<0.05

Control = 28% glycerol (10 ml/kg B. Wt.).

Values are expressed as mean ± S. E.

NS - Not Significant

Urine osmolality was not significantly different from the control group. Mean urine osmolality actually was lower in the denervated group. This might have been related to the slightly higher urine flow rates in the latter group.

Table III shows the effect of 28% I.M. glycerol on renal function in the rats subjected to prior unilateral denervation or Sham denervation with contralateral nephrectomies. All measured parameters of renal function showed marked improvements with prior denervation. This is illustrated graphically in Figure 1.

c. Adequacy of Denervation:

To determine the success of denervation, renal catecholamine contents were measured in 4 out of 12 randomly selected unilaterally denervated rats. The remaining 8 rats were subjected to intramuscular glycerol administration. Renal catecholamine contents in the remaining 8 rats were determined 48 hours post-glycerol administration.

Results are shown in Table IV. No significant differences in renal catecholamine contents were observed before or after glycerol treatment. Ten of the twelve denervated left kidneys contained no measurable catecholamines. The remaining 2 kidneys contained less than 12% of the catecholamines contained in their contralateral innervated control.

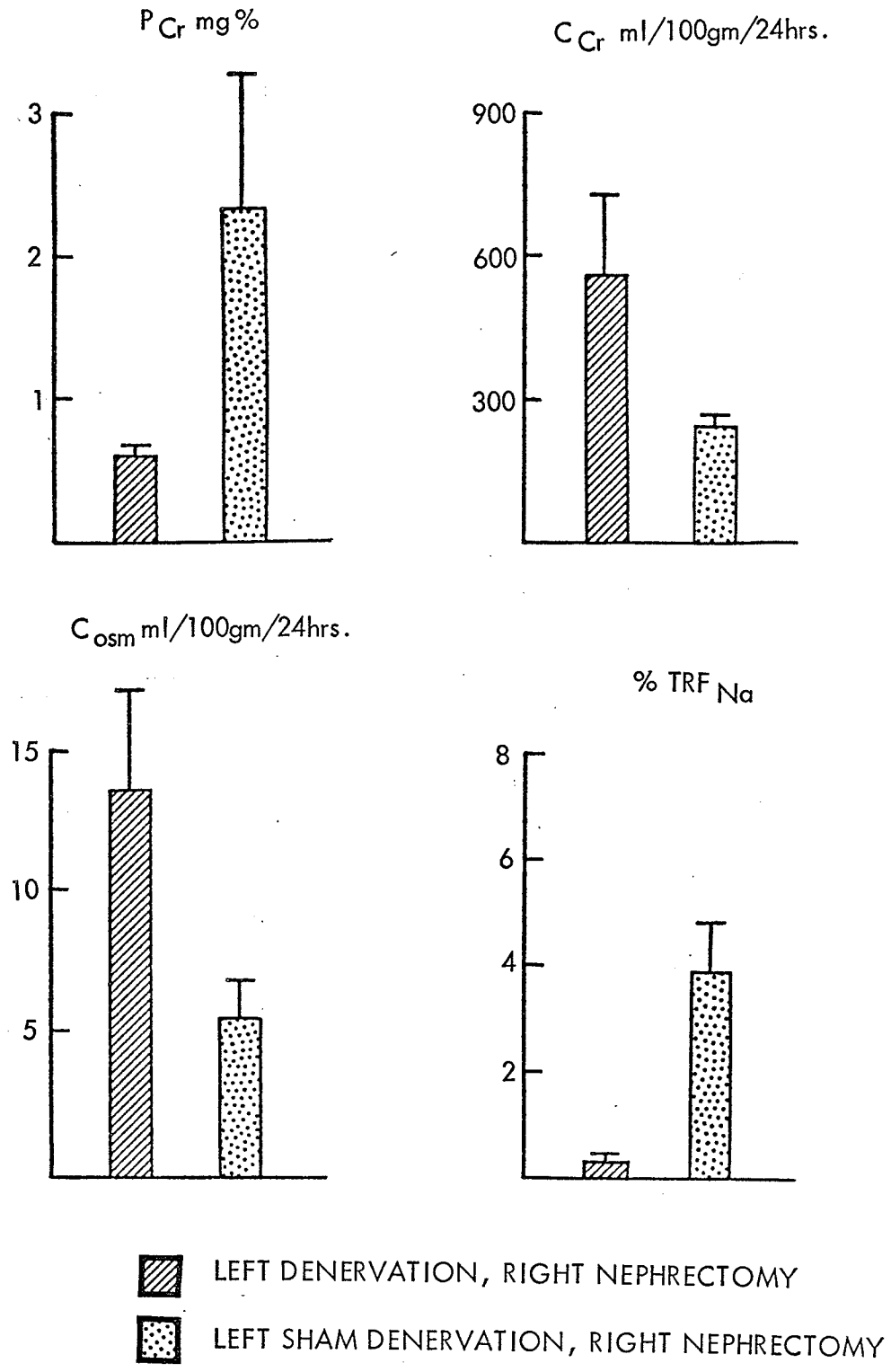


Figure 1: Effect of 28% glycerol on renal function in the rat: renal denervation versus sham denervation with contralateral nephrectomies.

TABLE III

Effect of Prior Denervation on Glycerol-induced Acute Renal Failure. Renal Function 24 to 48 Hours Post-glycerol Administration

	Left Denervation Right Nephrectomy (n=5)	Left Sham Denervation Right Nephrectomy (n=4)	p Value
$\dot{V}$ (ml/100 gm/24 hrs.)	9.1 ± 2.2	6.8 ± 3.1	NS
C_{Cr} (ml/100 gm/24 hrs.)	481.0 ± 136.4	56.14 ± 11.9	< 0.05
C_{osm} (ml/100 gm/24 hrs.)	13.5 ± 3.5	5.6 ± 1.2	< 0.05
C_{H_2O} (ml/100 gm/24 hrs.)	-4.4 ± 2.9	1.25 ± 1.16	< 0.01
U_{osm} (mOsm/kg)	1043.0 ± 201.8	638.0 ± 113.9	NS
% TRF_{Na} (%)	0.36 ± 0.29	3.8 ± 1.08	< 0.01
P_{Cr} (mg%)	0.65 ± 0.04	2.38 ± 0.92	< 0.05

TABLE IV

Effect of Denervation on Renal Catecholamine Content. Six of the Eight Denervated Left Kidneys in the Post-glycerol Group Showed No Detectable Catecholamines

	Left Kidney (Denervated) ug/gm	Right Kidney (Innervated) ug/gm	N	p Value
Before Glycerol Administration	0	0.085 ± 0.021	4	< 0.001
After Glycerol Administration	0.006 ± 0.004	0.118 ± 0.014	8	< 0.001

d. Comments:

It is quite obvious that prior renal denervation offered considerable protection against glycerol induced acute renal failure. The better preserved renal function 24 to 48 hours post-glycerol administration may represent either the production of a less severe lesion or an earlier recovery, since the occurence of diuresis appeared sooner than in the control group. It could be argued that the use of endogenous creatinine clearances may not reflect accurately glomerular filtration in the rat. In such cases, assuming a relatively constant daily production of endogenous creatinine, plasma creatinine concentration may actually be a better index of glomerular function. Nevertheless, the use of endogenous creatinine clearance under these experimental conditions may be qualitatively acceptable. Although it is true that the more severe the tubular lesion, the greater may be the extent of back diffusion, but the quantity of tubular creatinine secretion would be diminished as well. The two opposing effects on creatinine excretion thus tend to minimize the error introduced.

Protection against glycerol induced acute renal failure by prior renal denervation is consistent in the thesis that the sympathetic nervous system plays a role in the pathogenesis of the lesion.

Endogenous creatinine clearances and plasma creatinine concentrations in the unilaterally denervated rats (n = 4) were not signifi-

cantly different from normal rats (n = 20). However, one must safeguard against the all-too-committing statement of renal denervation with depletion of catecholamines being identical to sympathectomy alone, and thus conclude adrenergic mediation was responsible for the protection. Several lines of evidence have shown a relationship between the sympathetic nervous system, renal hemodynamics and renal sodium handling (Gill and Bantter, 1966; Schrier et al. 1967 and Bonjour et al. 1969). Renal renin content in denervated kidneys has been reported to be some 40% of innervated controls (Ueda et al. 1967).

Renal nerve stimulation or catecholamine infusion results in renin release (Vander, 1965; Bunag et al. 1966b). In denervated kidneys renin release is impaired (Ueda et al. 1967). In addition, denervation supersensitivity may occur and thus alter vascular smooth muscle reactivity. There may also be other unpredictable changes in other vasoactive systems concerned. Renal histamine content as shown in Table V is increased with denervation.

Thus, it is difficult to attribute conclusively the protective effect of chronic renal denervation due to hemodynamic improvement secondary to abolition of sympathetic overactivity. However, it does support the postulate that renal denervation is partially responsible for the observed protection from glycerol induced acute renal failure in the rat.

TABLE V

Effect of Denervation on Renal Histamine Content

	Left Kidney (Denervated) ug/gm	Right Kidney (Innervated) ug/gm	N	p Value
Before Glycerol Administration	4.74 ± 0.23	2.19 ± 0.10	4	< 0.005
After Glycerol Administration	5.48 ± 0.62	2.42 ± 0.28	8	< 0.001

3. Effect of Post-event Phenoxybenzamine on Glycerol-induced Acute Renal Failure

Since prior renal denervation offered partial protection in glycerol-induced acute renal failure, an attempt was made to determine if sympathetic blockade by phenoxybenzamine (POB) after glycerol administration would also modify the functional lesion. Protection by early post-event phenoxybenzamine blockade would support the thesis that considerable sympathetic overactivity is present early in the development of the lesion and this adverse overactivity is at least partly responsible for its pathogenesis.

a. Methods:

The same experimental protocol previously described was used with the following modification: Phenoxybenzamine at a dose of 2.0 mg/kg B.Wt. was administered intraperitoneally 2.5 hours after the administration of intramuscular glycerol. This dose of phenoxybenzamine when given intraperitoneally has previously been shown to be adequate in preventing systemic pressor responses to intravenous noradrenaline in the rat (McLean and Thomson, 1970).

After the collection of a 24-hour urine sample, animals were sacrificed 48 hours after glycerol injection. Renal function of the POB treated group was compared to the non POB treated controls.

b. Results:

Table VI shows parameters of renal function of the POB treated rats compared to controls. POB treated rats show significantly higher creatinine clearances, urine osmolality, and lower plasma creatinine concentrations as well as $\%TRF_{Na}$. Urine flow rates and osmolar clearances, however, did not differ significantly from non-POB treated rats.

c. Comments:

We have shown that phenoxybenzamine given 2.5 hours after glycerol injection can significantly reduce the severity of the resultant functional impairment 2 days later. It cannot be stated conclusively this is due to the alpha-adrenergic blocking activity alone. Phenoxybenzamine at higher doses is capable of blocking in vitro, and sometimes in vivo, the agonistic effects of other vasoactive substances including histamine, 5-hydroxytryptamine, acetylcholine and perhaps even angiotensin II (Nickerson, 1949; Nickerson and Hollenberg, 1967). Phenoxybenzamine may conceivably also inhibit the release of renin. It is also difficult to be certain how much of the phenoxybenzamine given intraperitoneally reached the renal vasculature in presence of severe renal ischemia as has been demonstrated by Ayers et al. (1971). Furthermore, the systemic administration of the drug makes it impossible to distinguish whether it is the systemic consequences of generalized alpha-adrenergic blockade or if it is the direct effect on the renal vasculature that

is significant in the production of the lesion.

However, the protective effect of post-event POB blockade and prior renal denervation compliment each other in supporting the hypothesis that the sympathetic nervous system plays a significant role.

TABLE VI

Effect of Intraperitoneal Phenoxybenzamine Administered 2.5 Hours Post-glycerol Injection. Renal Function at 24 to 48 Hours Post-glycerol Administration

	Control (n=20)	POB-Treated (n= 19)	p Value
$\dot{V}$ (ml/100 gm/24 hrs.)	9.7 ± 1.3	7.9 ± 1.0	NS
C_{Cr} (ml/100 gm/24 hrs.)	60.9 ± 21.6	221.8 ± 80.5	< 0.05
C_{osm} (ml/100 gm/24 hrs.)	9.4 ± 1.3	13.7 ± 1.9	NS
C_{H_2O} (ml/100 gm/24 hrs.)	13.8 ± 2.4	3.8 ± 1.8	< 0.05
U_{osm} (mOsm/kg)	426.1 ± 22.5	638.8 ± 83.8	< 0.05
% TRF_{Na} (%)	39.48 ± 11.49	8.04 ± 3.89	NS
P_{Cr} (mg%)	5.65 ± 0.65	1.69 ± 0.52	< 0.0005

Control 28% glycerol (10 ml/kg B.Wt.).

Values are expressed as means ± S.E.

NS - Not Significant

B. Noradrenalin-induced Acute Renal Failure in the Dog

The need for an experimental model of acute renal failure where serial hemodynamic measurements can be made and correlated with renal function has been discussed (Section I). Experiments on glycerol-induced acute renal failure in the rat have implicated that the sympathetic nervous system plays a role in the development of the lesion, and that this could be a vascular-mediated phenomenon.

To further substantiate this claim, it is necessary to demonstrate that sustained sympathetic overactivity can produce acute renal failure, and that this is associated with the pattern of change in renal perfusion characteristic of acute renal failure.

Clinical circumstances such as crush injuries, major burns, shock, hemorrhage, etc. under which acute oliguric renal failure develops are generally associated with increased sympathetic activity and elevated circulating catecholamine levels (Bull et al. 1950; Kiley et al. 1960 and Merrill, 1960). Although direct or indirect stimulation of renal nerves causes diminished renal blood flow as well as a redistribution of blood flow characterized by relative cortical hypoperfusion similar to the pattern seen in established acute renal failure (Pomeranz et al. 1968; Aukland, 1968b), tolerance develops following prolonged sympathetic stimulation (Block et al. 1952; Kubicek et al. 1954). The phenomenon of autoregulation appears to take over control and renal failure does not develop.

Catecholamines have been shown to have profound effects on the renal vasculature. Adrenaline or noradrenaline, when administered intravenously or intra-arterially, causes a decrease in total renal blood flow (Moyer and Handley, 1952; Spencer et al. 1964; Aviado et al. 1958; and Langston et al. 1962) as well as a pattern of intrarenal distribution of flow similar to that of sympathetic stimulation (Aukland, 1968b; Carriere, 1969). Less quantitative but more direct methods employed including fluorescent studies (Moses, 1952) and arteriography (Abrams et al. 1962; and Elkin and Meng, 1963) have also demonstrated cortical ischemia. Hatcher et al. reported in 1963 that adrenaline, when infused directly into the renal artery could produce irreversible renal failure in the dog. However, little is known regarding the renal hemodynamic or functional aspects of this model. Hollenberg (personal communications) also has found that prolonged intra-arterial infusion of noradrenaline can cause acute renal failure in the dog, associated with poor renal cortical perfusion. Thus, it appears that the agonistic properties of noradrenaline, the neurotransmitter of the adrenergic nervous system, may be utilized to circumvent the problem of tachyphylaxis following sustained sympathetic stimulation.

Therefore, we attempted to investigate the effects of prolonged intra-arterial infusion of noradrenaline on renal hemodynamics and renal function in the dog. The development of reproducible acute renal

failure would therefore provide us with an experimental model where direct renal hemodynamic measurements can be made and correlated with renal function. This would also allow us to investigate the possibility of a vascular-mediated lesion.

1. General Methods

(i) Animal Preparation, Experimental Setup and General Procedure.

Mongrel dogs of either sex weighing 12-30 kg free from obvious disease or malnutrition were used. Pregnant or lactating animals were rejected. The animals were housed in a constant temperature environment of approximately 68°F, and fed a standard laboratory purina chow pellets. Free access to water was available at all times. Food was withheld from animals 16-20 hours prior to experiment.

Under pentobarbital anesthesia (33 mg/kg B.Wt. given I.V.), the animals were intubated with the appropriate size of cuffed McGill endotracheal tubes and connected to a constant volume ventilator. All experiments were carried out under controlled ventilation on room air, with the environmental temperature approximately 68-70°F. Following the implantation of a non-cannulating electromagnetic flow probe (Statham, Type MDS), total renal blood flow of the right kidney was measured by a Statham square-wave electromagnetic flowmeter, Model 5000. Electromagnetically derived blood flows were calibrated frequently throughout the course of study in vitro, using heparinized whole blood and a segment of the common carotid artery in a saline bath.

Electronic-zero blood flow was checked frequently during each experiment, and checked against mechanical-zero blood flow at the conclusion of each experiment. Mean total renal blood flow was obtained by electronic integration.

All drugs to be administered intrarenally were given via a polyvinyl catheter (O. D. = 0.046 in.; I. D. = 0.028 in.) inserted into the right renal artery. The patency of the catheter was ensured by the constant infusion of 0.9% saline by a Harvard infusion-withdrawal pump at a fixed rate of 0.194 ml/min.

In acute experiments, the left brachial artery was cannulated and arterial blood pressure monitored throughout the experiment using a Statham P23A pressure transducer. Mechanical calibration of the pressure transducer was carried out frequently during the course of the entire study. The right internal jugular vein was cannulated and the polyethylene catheter advanced to the right atrium. This cannula served for blood sampling, central venous pressure monitoring and intravenous fluid and drug administration. Bipolar electrocardiogram was recorded throughout the experiment for the monitoring of cardiac rhythm. Using a left flank incision, the left ureter was approached retroperitoneally and cannulated for the collection of urine samples using a PE90 polyethylene catheter. A similar right flank incision was used for approaching the right ureter for cannulation. All ureteric cannulae had a dead space of 1.0 ml, and were advanced to the level

of the renal pelvis. The right renal artery was mobilized and a polyvinyl catheter was inserted using a modified technique of Herd and Barger (1964). Total renal blood flow measurements before and after the catheterization of the renal artery were identical. The duration of mechanical handling of the right renal artery for this purpose was usually less than 3 minutes. Animals with double or multiple right renal arteries or early bifurcations were rejected for experimentation. All experiments were performed in the left decubitus position.

Thirty minutes was allowed for stabilization after surgery, followed by a 1 hour control period. Blood and urine samples were obtained for conventional clearance studies. Noradrenaline at a dose of 0.4-4.0 ug/kg/min, calculated as the weight of the base, was infused intra-arterially to the right kidney via the polyvinyl catheter using a Harvard constant infusion pump. The infusion period lasted 2 hours. The rate of infusion of noradrenaline was in multiples of 9.7 ug/min, and was the rate required to keep right total renal blood flow at zero. This generally resulted in considerable spilling over of noradrenaline into the systemic circulation, resulting in systemic hypertension during the 2 hour infusion period. Further increase in the rate of noradrenaline infusion was always limited by the development of cardiac arrhythmias.

In all acute experiments, urine samples in 2 hour blocks were collected separately from both kidneys for conventional clearance

studies. Blood samples were taken in the middle of each clearance period for the determination of plasma creatinine concentration, Na^+ and K^+ concentrations, osmolality and hematocrit. Urine samples were analysed for creatinine concentration, Na^+ and K^+ concentrations and osmolality. Endogenous creatinine clearances (C_{Cr}), osmolar clearances (C_{osm}), free water production (C_{H2O}), % tubular rejection of filtered sodium load (% TRF_{Na}) were calculated by conventional formulae. Statistical analyses used were as indicated in the individual experiments.

In chronic experiments, the procedures outlined above were slightly modified. All surgery was performed under sterile conditions. Ureters were cannulated by teflon tip (No. 16) silastic cannulae, brought out through individual stab wounds in the back, and connected to closed drainage bags secured to the back of the animal. This allowed for accurate collection of urine volume from separate kidneys for the entire duration of the experiment. A chronic indwelling polyethylene central venous pressure catheter was placed through a cut-down of the right internal jugular vein for periodic blood specimen sampling. The catheter was filled with heparin to maintain patency and was protected by a neck plaster cast. Following a 2 hour infusion of noradrenaline, the animals were allowed to recover and renal function was followed at intervals. Renal hemodynamics were assessed by the $Xenon^{133}$ washout method (to be described later) and by selective renal angiography.

(ii) Intrarenal Blood Flow Distribution

Intrarenal blood flow distribution was assessed by a modified $Xenon^{133}$ washout method as described by Hollenberg et al. (1968). This consisted of a rapid intra-arterial injection of 0.3-0.7 millicurie of $Xenon^{133}$ dissolved in 0.5-1.0 ml of 0.9% saline followed immediately by 0.5-1.0 ml of 0.9% saline into the renal artery. In chronic experiments, injection of $Xenon^{133}$ was done through a catheter placed in the renal artery via the femoral artery by standard angiographic techniques under fluoroscopy.

The disappearance of radioactivity in the kidney was monitored externally by a scintillation probe containing a 2-inch NaI crystal placed over the kidney. The duration of counting was 3 minutes. The probe output was led through a pulse height analyzer with a window setting of 70 to 120 Kev (High Voltage Supply = 1350 volts) to a digital timer-rate-meter and then either to a direct printer or a tape recorder for subsequent off-line digital print out. Optimal settings for the pulse height analyzer were checked each time before the experiment.

Based on the theories proposed by Kety (1951) and described in detail by Thorburn et al. (1963), the rate of radioactivity washout from the kidney is a complex function of renal blood flow and can be described

in the normal canine kidney by the sum of four exponential functions:

$$A(t) = A_1 e^{-k_1 t} + A_2 e^{-k_2 t} + A_3 e^{-k_3 t} + A_4 e^{-k_4 t}$$

Where: $A(t)$ = Total Activity at time t

A_i = Initial Activity of compartment i

k_i = Rate constant for compartment i

t = Time

It has been shown that for the kidney under normal and certain experimental conditions, including drug induced ischemia and hemorrhagic hypotension, each individual exponential function corresponded to a certain particular anatomic region within the kidney by autoradiography (Thorburn et al. 1963; Carriere et al. 1966,1971; Carriere, 1969; Carriere and Friborg, 1969; and Fisher et al. 1970). The advantages, disadvantages and limitations of this method of assessment of intrarenal renal blood flow have been discussed by numerous authors (Ladefoged, 1966; Ladefoged and Pedersen, 1967; Andersen and Wise, 1969; and Grünfield et al. 1971). Briefly, there is good correlation between blood flow obtained by electromagnetic flow meters, direct renal venous outflow and the inert gas desaturation method (Ladefoged and Pedersen, 1967; Ladefoged and Fritjossan, 1968; and Bell and Harper, 1968). Although it is impossible to identify individual compartments when the difference in their corresponding time constants is less

than 3 times (Dobson and Warner, 1957; Riggs, 1963), it is generally agreed that in the normal kidney, the fast flowing component I represents cortical blood flow; component II, III and IV represent corticomedullary, inner medullary flow and flow through pericapsular and perihilar fat respectively.

From the slopes and intercepts of components I, II and III + IV, blood flow in ml/100 gm of tissue/min and percentage of total blood flow can be computed. It has been shown by Rosen et al. (1968) that 3 minute-washout curves are sufficient to yield blood flow information of the two faster flowing components. This method of analysis was used throughout the study. Mean renal blood flows where applicable were derived from the initial slopes of the composite washout curve (Ingvar and Lassen, 1962). All graphic analysis were carried out in duplicate at different times by the same individual (the author) from coded original data in order to minimize bias and variation. The means of the two graphical analyses of the same data were used in statistical analysis. Correction for the partition coefficient of Xenon between blood and tissue due to variations in hematocrit were made according to the equation suggested by Andersen and Ladefoged (1969).

All $Xenon^{133}$ washout curves were recorded with the animals under pentobarbital anesthesia.

(iii) Selective Renal Angiography

Renal angiography was performed in selected experiments under pentobarbital anesthesia in conjunction with $Xenon^{133}$ washout studies. It has been shown by Hollenberg et al. (1968c) that contrast agents such as sodium and methylglucamine diatrizoate had no appreciable effect on renal hemodynamics.

Arterial catheterization was carried out by an experienced angiographer (I.M.). Using the Seldinger technique or a direct femoral arterial cut down under fluoroscopic control, 5-10 ml of Renografin-60^{R} (Meglumine and sodium diatrizoate 60%) was injected rapidly into the renal artery. Serial films were taken on a Schonander cut film changer. Kidney size, presence, diameter and regularity of filling of the renal arterial vasculature were examined. The nephrogram, transit time and reflux of dye into the aorta were qualitatively assessed.

(iv) Plasma Renin Activity

Plasma renin activity in selected experiments in the dog were measured by a modified radioimmunoassay Haber method (Haber et al. 1965). Lower limit of sensitivity by this method is 0.3 ng Angiotensin I generated/ml/hr.

Briefly, anti-angiotensin, produced in rabbits following administration of angiotensin-poly-L-lysine polymer, was allowed to react with labeled angiotensin. Upon addition of unlabeled angiotensin to

this mixture, some of the labeled angiotensin was displaced from the complex and a quantitative relationship could be obtained between the amount of unlabeled angiotensin, and labeled angiotensin released from the complex. Millimicrogram quantities of angiotensin could be determined. Reproducibility was approximately 15%.

(v) Histology

Renal tissues were (post-mortem, or precutaneous needle biopsy) fixed in either 10% formalin or formyl sublimate and were prepared for routine staining for light microscopy. Independent interpretation of histological abnormalities were documented independently by two individuals (AET and GH) in addition to the author.

2. The Effect of Intra-arterial Infusion of Noradrenaline on Renal Hemodynamics and Renal Function: The Model

(i) Chronic Experiments

These experiments were designed to investigate the long term effects of intra-arterial noradrenaline on renal hemodynamics and function in the dog, and, to determine whether prolonged intra-arterial noradrenaline could produce a lesion functionally and histologically similar to that seen in human acute failure.

a. Methods:

The general procedure has been outlined in the previous section. In this set of experiments 6 mongrel dogs of either sex, weighing from 13-27 kg were used. Following a 2 hour period of infusion of noradrenaline into the right renal artery, the animals were allowed to recover and their daily renal function followed. Twenty-four-hour urine samples from separate kidneys were collected from closed drainage bags and daily blood samples were taken via the chronic indwelling central venous pressure catheter. At periodic intervals, depending the state of recovery of renal function, bilateral selective renal angiography as well as assessment of intrarenal blood flow distribution, by the $Xenon^{133}$ washout technique, were carried out in most animals. Percutaneous renal biopsies were performed in C-1 and C-8.

In animals C-7 and C-8, intra-arterial infusion of noradrenaline was achieved by angiographic techniques as described; rather than by

surgical exposure of the renal artery. All angiographic investigations and Xenon^{133} washout studies for assessment of intrarenal blood flow distribution were performed under pentobarbital anesthesia and strict asepsis was observed.

b. Results:

All animals showed zero total right renal blood flow as measured by electromagnetic flowmeters during the 2 hour period of noradrenaline infusion. This was associated with systemic arterial hypertension. Complete anuria was observed in all cases in the infused kidney. A summary of the changes in renal function post-noradrenaline infusion is shown in Table VII. Termination of an experiment was usually due to the development of technical complications such as dislodgement or animal-induced breakage of the ureteral cannulae rather than due to infection or other illnesses. The duration of experiments ranged from 3 to 20 days. Following noradrenaline infusion, all animals showed signs of unequivocal functional acute renal failure in the infused kidney as compared to its contralateral control. This was shown by the development of a variable period of oligo-anuria lasting from a few hours to 3 days, marked depression of endogenous creatinine clearance, poor concentrating ability, an increased $\%TRF_{Na}$ and a high U_k/U_{Na} ratio when compared to the contralateral kidney. Due to the different rates of functional recovery in different animals, only the values obtained one day post-noradrenaline infusion and those obtained when

TABLE VII

Effect of Intra-arterial Noradrenaline Infusion on
Renal Function in the Dog: Chronic Experiments

Renal Function	Kidney	C-1		C-4		C-5		C-6		C-7		C-8	
		Low	High	Low	High	Low	High	Low	High	Low	High	Low	High
C_{Cr} (ml/min)	Right	0	5.94	0.5	3.2	11.2	23.0	9.1	13.9	7.92	8.2	0.49	3.12
	Left	-	-	48.7	32.2	49.4	38.4	24.3	27.3	13.3	16.3	16.3	14.9
V (ml/hr/kg)	Right	0	0.46	0.07	0.18	0.33	0.19	0.23	0.39	1.03	0.82	0.04	0.61
% TRF_{Na}	Right	0	2.76	1.18	0.93	0.84	0.01	0.5	0.85	1.71	2.5	1.38	4.72
U_{osm} (mOsm/kg)	Right	0	466	304	311	724	1730	1100	1224	603	675	732	476
	Left	-	-	432	542	1430	1568	2800	2160	1026	1500	1484	597
C_{H_2O}	Right	0	-1.55	0.09	-0.06	-0.95	-0.83	-1.49	-1.71	6.15	0.27	-2.52	-2.24
(ml/min/100 ml C_{Cr})	Left	-	-	-0.72	-1.20	-1.07	-1.00	-1.52	-1.82	3.98	3.12	-3.14	-1.61
Duration DAYS		10		9		8		15		1.5		20	

Low = value one day post-noradrenaline infusion to the right kidney.
High = value obtained when highest C_{Cr} of right kidney was achieved during the course of functional recovery.

maximum creatinine clearance had been achieved during recovery are presented in Table VII.

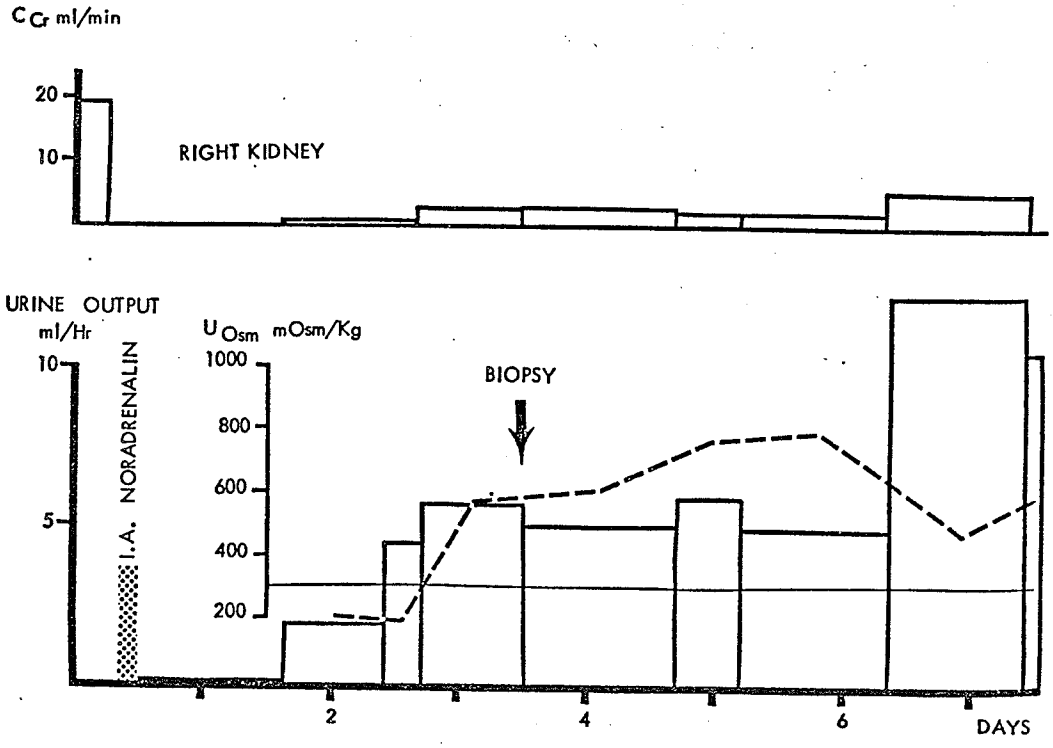
All animals showed incomplete return of renal function associated with return of urine flow rate. Some animals show a diuretic phase. Although $U/_{Posm}$ ratios for the experimental right kidney were generally higher than one, they were lower than those of the contralateral left kidney at all times. Calculated values for free water clearances per 100 ml of glomerular filtrate of the experimental organs always exceeded those of the contralateral side. $\%TRF_{Na}$ for the infused kidney were characteristically high, and $\frac{U_K}{U_{Na}}$ ratio characteristically low as seen in human acute renal failure. Figure 2 shows the functional course of a typical chronic experiment, demonstrating the initial oliguria, depressed endogenous C_{Cr}, hypo-osmotic urine with subsequent progressive improvement.

Histological studies of autopsy or biopsied material by light microscopy revealed variable tubular necrosis, debris and casts within tubular lumena, and evidence of regeneration (tubular epithelial cell mitoses). Glomeruli and arterioles showed no obvious histologic changes. Scattered patchy areas of infarct and hemorrhage were sometimes seen. A typical histological picture is shown in Figure 3.

Selective renal angiography performed in 6 animals (C-4, C-5, C-6, C-7, C-8, C-1) at various stages of renal failure demonstrated abnormalities in the right renal vasculature characterized by irregular

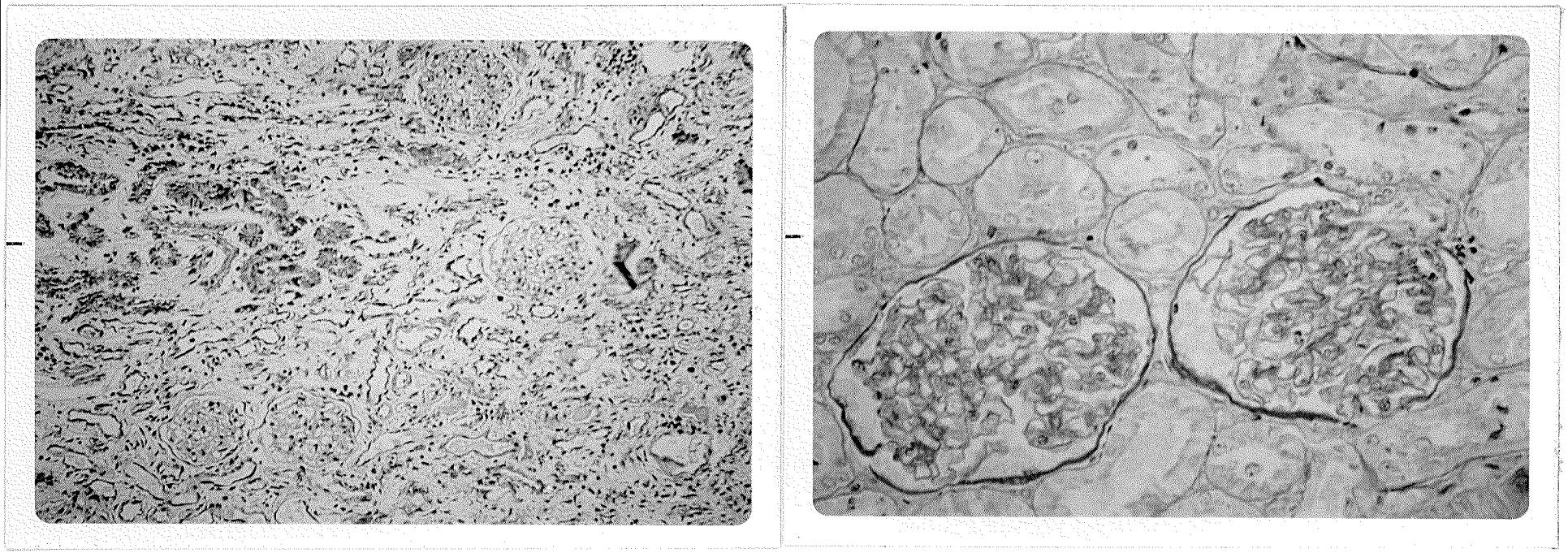
outlines of first, second and third order vessels, diminished penetration of Renografin^{R}-60 into the cortical region and poor or non-visualization of the higher order vessels. The most marked disturbances in angiographic appearance of the renal vasculature occured shortly after the infusion of noradrenaline, and, these became less evident as renal function subsequently improved. Figure 4 shows the serial right selective renal angiograms of experiment C-8. Prior to noradrenaline, the renal vasculature appeared normal (Figure 4A). Twenty minutes after completion of the 2 hour noradrenaline infusion, there was no visualization of the renal vasculature beyond the first major branches (Figure 4B). Twenty-four hours post-noradrenaline infusion, renal cortical vasculature was again visualized. By 19 days post-noradrenaline infusion, the angiographic appearance was only slightly different from control. Serial angiographic studies of the contralateral side (not shown) remained within normal limits at all times throughout the study.

Intrarenal blood flow distribution post-noradrenaline infusion as assessed by the Xenon^{133} washout technique, showed a significant decrease in the percentage of blood flowing to the first component C_I (outer cortex), with a reciprocal increase in percentage of blood flowing to the second component C_{II} as compared to the pre-infusion control. Flow rates expressed in ml/100 gm/min to these two compartments



EXP. C-1

Figure 2: Time course of renal function in noradrenaline-induced acute renal failure in the dog: chronic experiment (C-1).



A. LOW POWER (X10)

B. HIGH POWER (X25)

Figure 3: Histological appearance of right kidney in noradrenaline-induced acute renal failure (Exp. C-8), 19 days post-infusion. Patchy tubular necrosis and regeneration are evident. Debris and casts are present within lumina. Glomeruli and vessels do not show any obvious histologic change.

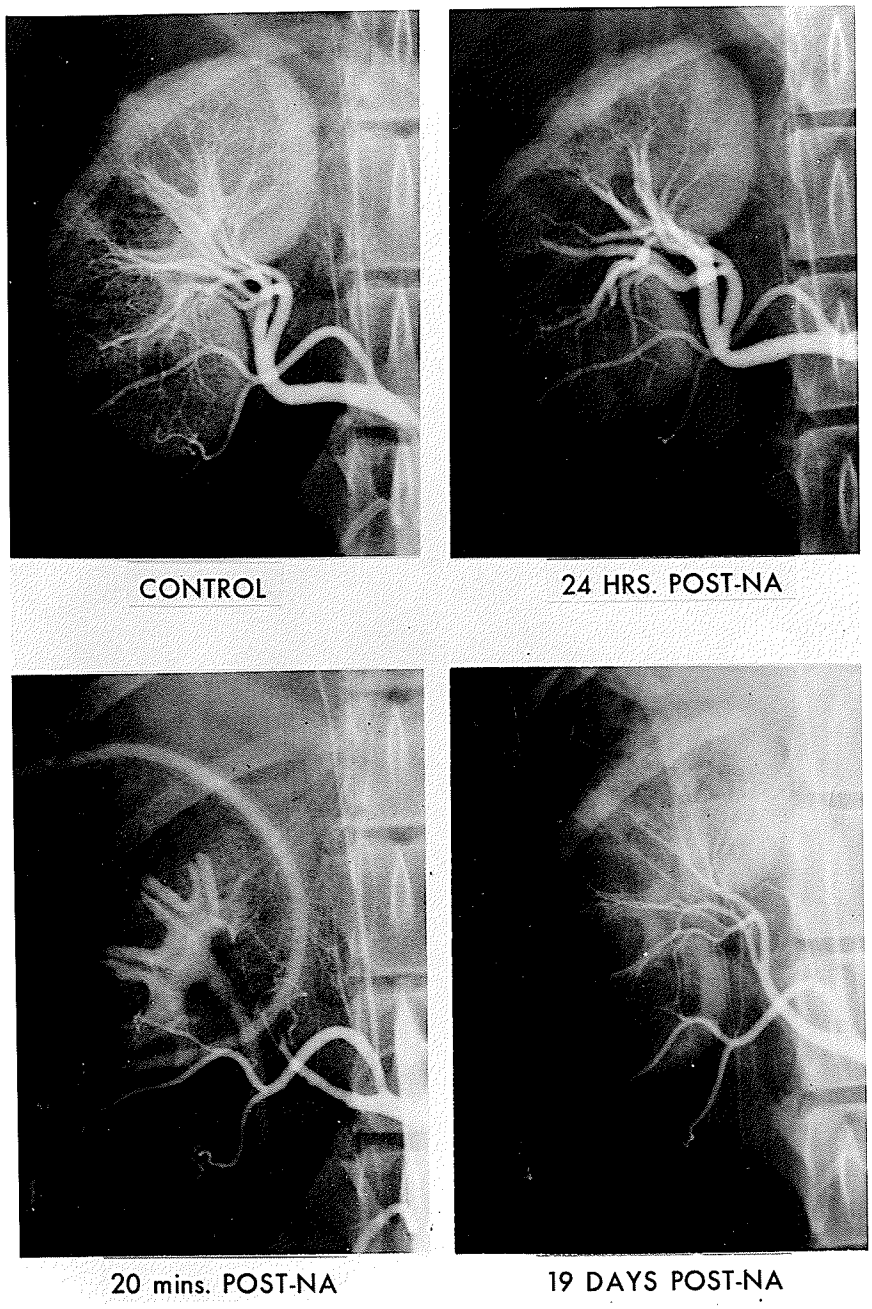


Figure 4: Angiographic appearance of right kidney in noradrenaline-induced acute renal failure (Exp. C-8).
4A - prior to noradrenaline infusion normal angiogram; 4B - 20 minutes post-infusion, no contrast material beyond first major branches; 4C - 24 hours, and 4D - 19 days post-infusion, both showing sustained reduction in cortical perfusion.

exhibited more variable changes. Mean renal blood flow was reduced in all post-noradrenaline infused kidneys. The results of IRBD are shown in Table VIII and a typical set of Xenon^{133} washout curves for experiment C-8 is shown in Figure 5.

Urine cultures remained sterile during the entire experimental period.

c. Comments:

The foregoing chronic experiments have demonstrated that, with infusion of noradrenaline directly into the renal artery for a 2 hour period, one could induce unequivocal functional acute renal failure. The course of this lesion was characterized by an initial period of oligo-anuria, depressed glomerular function, impaired tubular function, and cortical underperfusion. Histologically, the lesion was that of acute tubular necrosis rather than cortical necrosis and infarction. Qualitative changes observed in this experimental model and the propensity to recover are remarkably similar to the acute renal failure seen in man.

It can be concluded that prolonged I. A. noradrenaline can induce potentially reversible unilateral acute renal failure in the dog. The animals showed no signs of ill health due to the normally functioning contralateral kidney. Plasma creatinine concentrations, osmolality and electrolytes were all within normal limits. No evidence of urinary

TABLE VIII

Effect of intra-arterial infusion of noradrenaline on IRBD in the dog: chronic experiments

	RBF ml/100 gm/min			% RBF		
	C_I	C_{II}	Mean	C_I	C_{II}	$C_{III + IV}$
Normal Values (N=11)	425.3 (34.3)	96.5 (4.2)	312.9 (26.2)	75.6 (1.8)	19.8 (1.8)	4.7 (0.4)
C-4 (4 days Post-NA)	391.6	87.0	130.54	30.5	48.8	19.4
C-5 (8 days Post-NA)	535.5	95.9	139.7	36.5	49.9	14.5
C-8 (19 days Post-NA)	671.1	98.7	127.1	26.7	52.1	21.1
Mean (S.E.) N=3	532.7 (80.7	93.9 (3.5)		31.2** (2.9)	50.3*** (1.0)	18.4*** (2.0)
C-6 (1 day Post-NA)	459.0		128.5	38.4		61.6
C-7 (1 day Post-NA)	160.8		144.7	87.3		12.7
Mean (S.E.) N=5			143.1** (7.8)			

Rapid-flow components (C_I) could not be identified in 2 of the 5 Xenon ^{133} washout curves performed early (1 day) after noradrenaline infusion.
Non-paired t test used for statistical analysis.
* P <0.05 ** P <0.01 *** P <0.001
Results without p Values = No significant difference from normal.

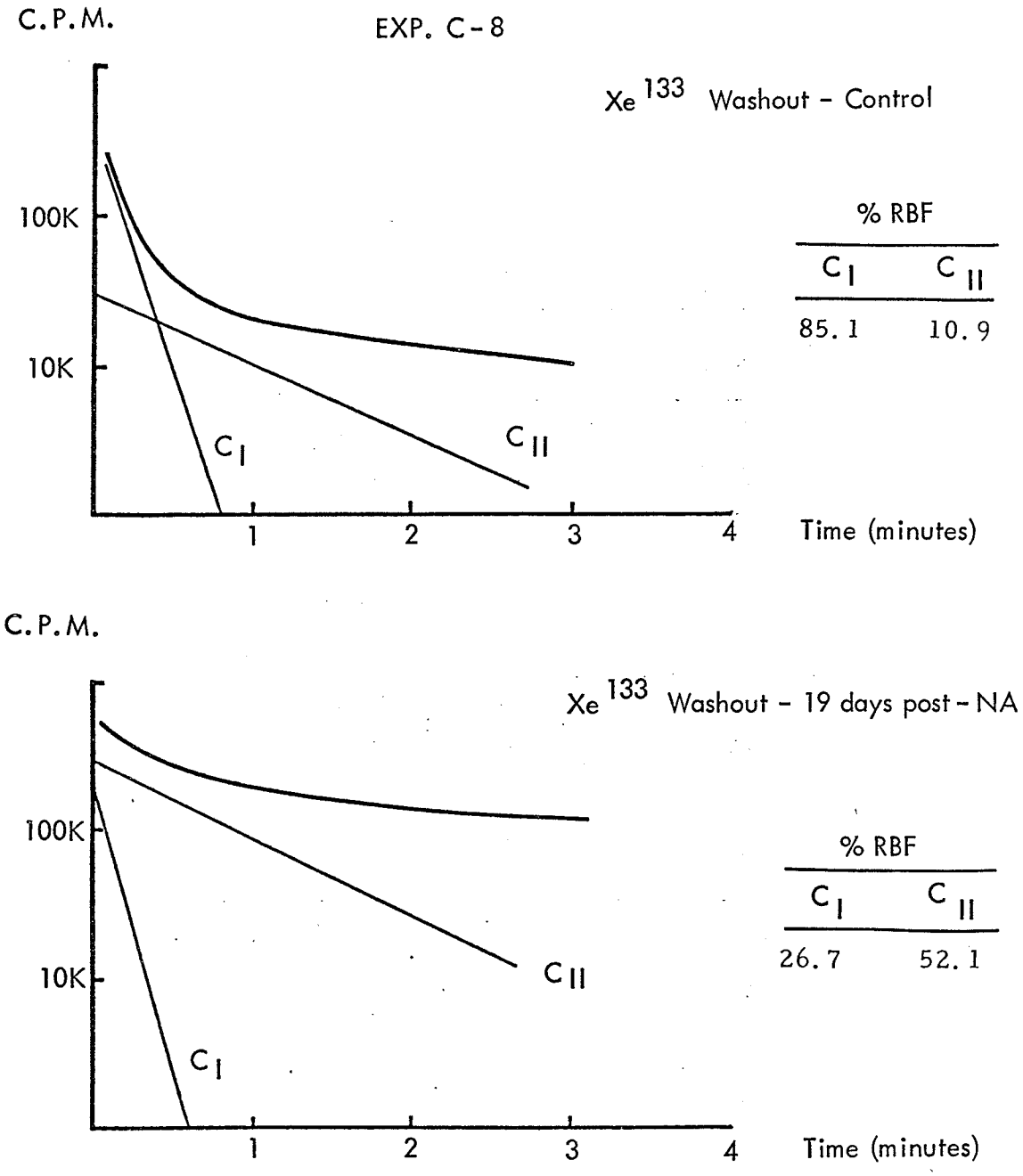


Figure 5: IRBD in noradrenaline-induced acute renal failure in the dog: chronic experiments.

tract infection was found. Thus, it appears that noradrenaline-induced acute renal failure in the dog may prove to be a satisfactory model for studying the role of vascular mechanisms in the pathogenesis and persistence of the lesion as outlined in Section I. The presence of an intact contralateral kidney serves to minimize or prevent possible contributions from secondary effects of impaired function per se.

(ii) Acute Experiments

Since it was possible to induce acute renal failure by prolonged intra-arterial infusion of noradrenaline, an attempt was made to document and investigate the hemodynamic events leading to and associated with the development of acute renal failure and to investigate renal function in early established renal failure.

a. Methods:

The general experimental procedure is as described in Section III, B. Nine mongrel dogs of either sex, weighing 12-28 kgs were used. As mentioned before, the rate of infusion of noradrenaline was the dose required to keep total renal blood flow to the right kidney at zero as measured by an electromagnetic flowmeter. Pentobarbital was supplemented intravenously when necessary during the course of the experiment. Urine samples were collected separately from both kidneys for a 1 hour control period, a 2 hour noradrenaline-infusion period and 2 hourly thereafter for 6 additional hours.

b. Results:

The effects of intra-arterial infusion of noradrenaline on renal hemodynamics are shown in Tables IX and X. As can be seen in Table IX, during the infusion of noradrenaline to the right kidney, total right renal blood was kept at zero while systemic arterial blood pressure rose markedly. Occasional cardiac irregularities, usually bigeminal rhythm were noted. This either subsided spontaneously or with decreasing the rate of noradrenaline infusion. Systemic arterial blood pressure returned to control values with cessation of noradrenaline infusion. Right total renal blood flow remained severely depressed. The rate of return towards control values showed considerable variation from animal to animal, and, by the end of 6 hours, mean total renal blood flow was approximately 50% of control; mean right renal vascular resistances was correspondingly elevated.

Intrarenal blood flow distribution, as assessed by $Xenon^{133}$ washout, following noradrenaline infusion is shown in Table X. While the fast flow component C_I was present during the control period, it could not be identified up to 6 hours post-infusion. Mean RBF was also reduced.

The effect of intra-arterial infusion of noradrenaline on renal function is shown in Table XI. The right kidney was anuric during noradrenaline infusion in all experiments. In 4 out of 9 experiments, the

right kidney remained completely anuric for the subsequent 6 hour period, although total renal blood flow had shown considerable recovery. There was no statistical difference in total renal blood flow 6 hours post-noradrenaline infusion between the anuric and the non-anuric animals. Regardless of the urine flow rate, endogenous creatinine clearances were all markedly depressed. As well, values for $\%TRF_{Na}$, U_{osm}, U_k/U_{Na} were all typical of established acute renal failure. Left renal function was only transiently affected by the infusion of noradrenaline. These essential features are diagramatically shown in Figure 6.

Histologically, acute tubular necrosis was observed in the right kidney. The microscopic appearance of a typical experiment (Exp. 103) is shown in Figure 7.

c. Comments:

These acute experiments again demonstrated that intra-arterial noradrenaline given over a 2 hour period, could induce acute renal failure. This was associated with very dramatic vascular events, both systemic and local. Evidence of gross impairment in function was present immediately following cessation of noradrenaline infusion. There was an associated renal hypoperfusion due to a persistence of increased renal vascular resistance even in absence of continued exogenous noradrenaline. The increase in resistance resides mainly in the cortex. The post-noradrenaline depression in GFR and other

modalities of function persisted in the presence of increases in total renal blood flow which reached or exceeded 50% of control during the 6 hour period of observation.

In the noradrenaline-dog model, in contrast to the human situation where precipitating events are frequently long continuing, the early transient phase characterized by increased sodium reabsorption and potential reversibility, was not documented.

The systemic effects of markedly elevated arterial blood pressure, the activation of the sympathetic nervous system and other vasoactive systems due to the escape of noradrenaline into the systemic circulation made it difficult to attribute the induction of acute renal failure by noradrenaline purely due to its local effects on the kidney. Sympathetic activation is a potent stimulus to the release of renin and vasopressin (Vander, 1965; Bunag et al. 1966a, 1966b). Also prolonged intravenous noradrenaline infusion has been shown to produce plasma volume depletion, shock (Sutter, 1963), and to accelerate the process of vascular decompensation (Hollenberg, 1965). Central venous hematocrits, measured at 2 hourly intervals in the present study, invariably showed significant increases during and immediately after noradrenaline infusion from values of 40% to as high as 60%. There was a slow return towards control values by 6 hours. Even with these changes, the contralateral kidney showed minimal or no impairment in function.

TABLE IX

Effect of Intra-arterial Infusion of Noradrenaline on Renal Hemodynamics in the Dog: Acute Experiments
N=9

Right Kidney	Control	NA Infusion	2 Hrs. Post-NA	4 Hrs. Post-NA	6 Hrs. Post-NA
RBF (% Control)	100	1.9*** (1.4)	53.9*** (8.3)	70.2*** (4.8)	69.6*** (10.1)
Mean B.P. (mm Hg)	129.4 (4.7)	168.9** (9.1)	126.7 (6.3)	122.2 (4.5)	119.4 (5.4)
RVR (% Control)	100	8***	217.5** (40.0)	140.4* (15.0)	131.7* (10.8)

* $P < 0.05$
** $P < 0.01$
*** $P < 0.001$
Paired t test was used for statistical analysis.
Values expressed as Mean (S.E.)
Values without p Values attached = No significant difference from control.

TABLE X

Effect of intra-arterial noradrenaline infusion on IRBD in the dog: chronic experiments

N=6

% RBF	Control	2 Hrs. Post-NA	p	6 Hrs. Post-NA	p
C_I	74.4 (2.9)	60.71 (2.9)		28.5 (1.5)	<0.001
C_{II}	21.2 (2.8)			58.2 (2.6)	<0.001
$C_{III + IV}$	4.3 (0.6)	39.3 (2.1)	<0.001	13.0 (0.7)	<0.001

Results expressed as Mean (S.E.).
Paired t-test was used for statistical analysis.

TABLE XI

Effect of Intra-arterial Noradrenaline Infusion on Renal Function in the Dog. Acute Experiments
N=9

Right Kidney	Control	NA Infusion	2 Hrs. Post-NA	4 Hrs. Post-NA	6 Hrs. Post-NA
$\dot{V}$ (ml/hr/kg)	1.10 (0.19)	0.03*** (0.02)	0.32*** (0.15)	0.39*** (0.18)	0.47*** (0.23)
$\frac{\dot{V}\text{ (Right)}}{\dot{V}\text{ (Left)}}$	0.84 (0.18)	0.02*** (0.12)	0.75 (0.31)	0.88 (0.43)	0.90 (0.47)
No. of dogs remaining anuric	0	9	4	4	4
C_{Cr} (% Control) Right	100	0***	3.6*** (1.9)	114.7*** (11.7)	12.7*** (8.2)
Left	100	103 (23.4)	98.0 (17.8)	103.8 (15.4)	90.7 (7.14)
U_{osm} (Right)	1.12	0.06***	0.31**	0.29**	0.30**
$\frac{U_{osm}\text{ (Right)}}{U_{osm}\text{ (Left}}$	(0.07)	(0.06)	(0.10)	(0.10)	(0.11)
% TRF_{Na}	1.33 (0.29)	0.60 (0.41)	11.82** (5.0)	19.64** (9.2)	15.2** (6.3)
U_K	0.11	0.04*	0.19	0.13	0.17
$\frac{U_K}{U_{Na}}$	(0.14)	(0.03)	(0.08)	(0.06)	(0.08)

Results expressed as mean (S. E.)
Paired t test was used for statistical analysis.
* P <0.05
** P <0.01
*** P <0.001
Values without p Values = No significant difference from control.

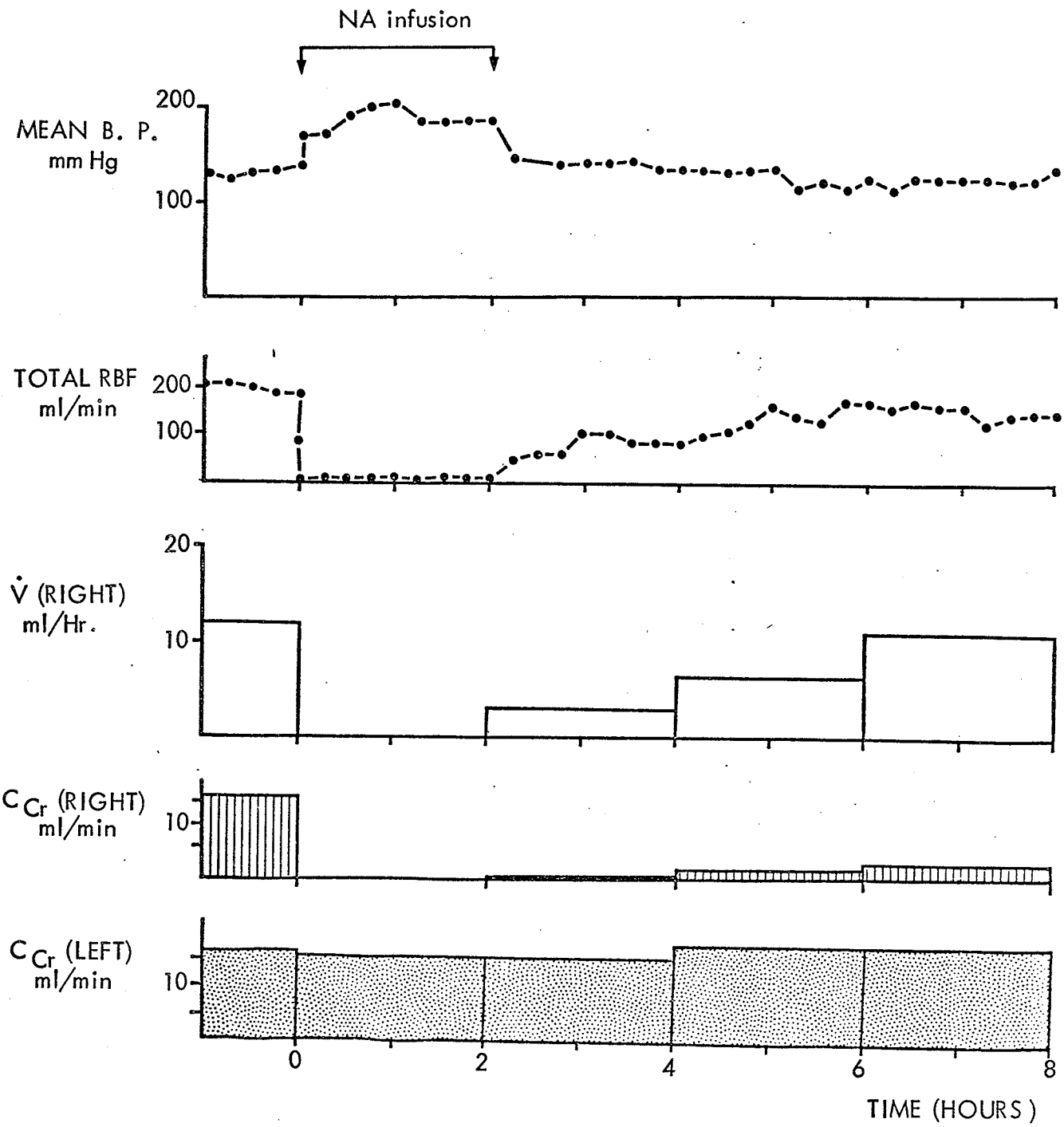


Figure 6: Renal hemodynamics and renal function following intra-arterial noradrenaline infusion in the dog.

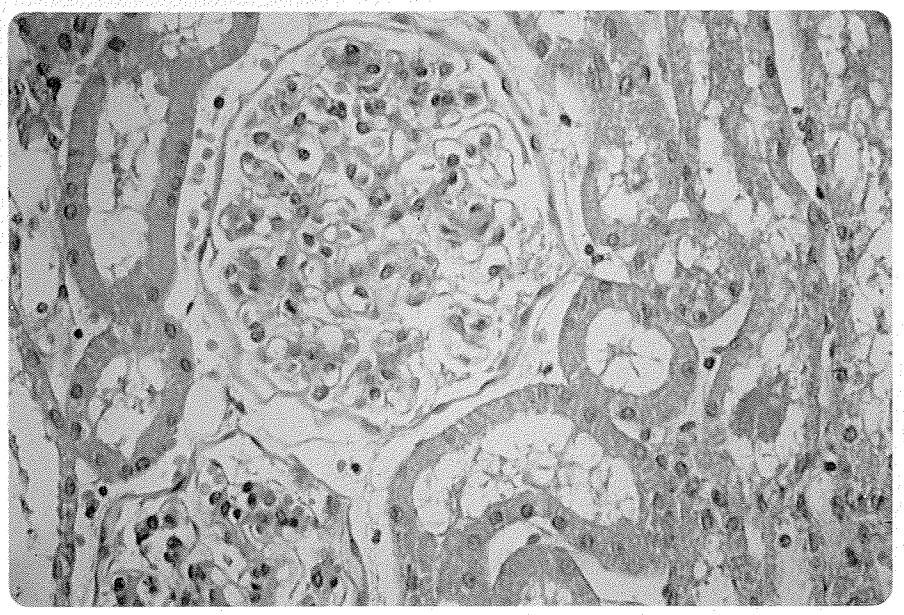


Figure 7: Histologic appearance of right kidney 6 hours post-noradrenaline infusion (Exp. 103). Glomerulus appears normal. Tubular necrosis and intraluminal debris can be seen.

3. Effect of Intravenous Infusion of Noradrenaline

In an attempt to exclude, in so far as possible, non-renal effects of the experimental procedure, noradrenaline was given intravenously in the same dose to a further group of animals.

a. Methods:

The general procedure was as described in Section III, B with the following modifications: Four mongrel dogs were used. Instead of infusing noradrenaline into the right renal artery, 0.9% saline was given at a rate of 0.194 ml/min. Noradrenaline, at a rate sufficient to achieve the same level of systemic hypertension as observed during the intrarenal artery administration was given intravenously for a 2 hour period. Again, at this rate of infusion, the occasional occurence of cardiac arrhythmias could be observed.

b. Results:

The effects of intravenous noradrenaline infusion for a 2 hour period in 4 animals are shown in Table XII. The rate of noradrenaline infusion required to produce comparable rises in systemic arterial pressure was the same whether administered into the renal artery or given intravenously.

There was a transient increase in calculated renal vascular resistance during intravenous noradrenaline infusion. At 2, 4 and 6 hours post-infusion, systemic arterial pressure, creatinine clearance, total renal blood flow and calculated renal vascular resistance did not

TABLE XII

Effect of Intravenous Noradrenaline Infusion on Renal Hemodynamics and Renal Function in the Dog
N=4

Right Kidney	Control	I. V. NA Infusion	2 Hrs. Post-NA	4 Hrs. Post-NA	6 Hrs. Post-NA
RBF (% Control)	100	88.0 (21.3)	77.1 (14.3)	79.8 (12.7)	93.0 (16.8)
Mean B. P. (mm Hg)	112.3 (8.3)	149.4 * (2.1)	100.0 (13.4)	98.1 (12.9)	100.0 (10.2)
RVR (% Control)	100	184.7 * (43.6)	124.2 (19.2)	113.7 (9.6)	103.3 (13.0)
C_{Cr} (% Control)	100	83.8 (20.4)	97.6 (17.1)	152.9 (75.1)	85.8 (6.9)
$\dot{V}$ (% Control)	100	41.9 * (14.2)	31.6 * (9.3)	44.4 * (9.2)	71.2 (21.3)
% TRF_{Na}	3.6 (1.1)	1.68 * (0.64)	0.89 * (0.49)	1.17 * (0.32)	2.48 (1.19)
$\frac{U_{osm}\,(Right)}{U_{osm}\,(Left)}$	0.95 (0.02)	0.97 (0.01)	0.95 (0.06)	0.96 (0.04)	1.05 (0.05)
C_{osm} (% Control)	100	33.6 * (17.2)	38.1 * (10.1)	47.9 * (11.3)	66.9 (12.2)

Values are expressed as mean (S. E.).

Paired t test was used for statistical analysis.

* $P < 0.05$

Values without p Values attached = No significant difference from control.

differ significantly from pre-infusion values. There was no evidence of persisting renal ischemia or of acute renal failure.

c. Comments:

It can be concluded that the effects of systemic noradrenaline together with the operative procedure of renal arterial catherization could not be responsible for the production of noradrenaline-induced acute renal failure in the dog.

4. Effect of Prior Phenoxybenzamine Blockade

Catecholamines have been known to have numerous poorly defined non-alpha and non-beta adrenergic effects on the cell (Innes and Nickerson, 1970). Parenteral infusion of catecholamines has been shown to result in myocardial lesions even when the coronary vasculature was practically devoid of alpha-adrenergic receptors (Moss and Schenk, 1970). Since a variety of nephrotoxins are capable of producing acute tubular necrosis in man and in experimental models, it is necessary to determine whether the induction of acute renal failure depends on alpha-agonistic activity and its vascular consequences or on other mechanisms. An attempt was made to study the effect of prior intra-arterial phenoxybenzamine followed by a 2 hour infusion of noradrenaline at the usual rate.

a. Method:

The general procedure was as outlined in Section III, B with the following modifications: Four mongrel dogs of either sex, weighing 14-22 kg were used. Following stabilization and a 30 minute control period, a small bolus dose of noradrenaline was given intra-arterially to demonstrate the vasoconstricting effect of noradrenaline. Phenoxybenzamine at a dose of 0.2 mg/kg B.Wt. (calculated as the base) was then slowly infused directly into the right renal artery over 10-15 minutes by a Harvard infusion-withdrawal pump. Noradrenaline was given for a 2 hour period at a rate sufficient to produce the same proportionate rise in systemic pressure as observed during the standard protocol. Serial urine and blood samples were obtained for the first 6 hours post-infusion.

b. Results:

Table XIII shows the effect of intra-arterial noradrenaline on renal hemodynamics and renal function in the kidney pre-treated with phenoxybenzamine. Following intra-arterial administration of phenoxybenzamine (0.2 mg/kg B.Wt.) to the right kidney, both urine flow rate and $\%TRF_{Na}$ increased significantly without much change in endogenous creatinine clearance, total renal blood flow, mean B.P. and renal vascular resistance. With subsequent intra-arterial noradrenaline infusion to the right kidney, mean arterial B.P. rose significantly

without concomittant increase in total right renal blood flow, indicating a decrease in renal vascular resistance. Endogenous creatinine clearance and urine flow rate showed slight transient decreases with gradual return to control values by 6 hours post-NA infusion. No oliguria or evidence of impairment of renal salt and water handling were observed. A typical experiment is graphically shown in Figure 8. Table XIV shows a comparison between the POB pretreated group (n = 4) compared to the non-POB treated group (n = 9). It is quite apparent the renal hemodynamics and renal function during and 6 hours post-noradrenaline infusion were well maintained in the POB pretreated group. Figure 9 shows the histological appearance of a POB pretreated kidney. Minimal evidence of tubular necrosis was seen. However, glomeruli and vessels showed no obvious histological lesions.

c. Comments:

From the above data, one can conclude that prior alpha-adrenergic blockade of the renal vascular bed by intra-arterial administration of phenoxybenzamine at a dose of 0.2 mg/kg B.Wt. completely prevented noradrenaline-induced renal ischemia and subsequent acute renal failure. Both renal hemodynamics and renal function were well preserved in the POB pretreated group. This dose of phenoxybenzamine was insufficient to produce significant systemic sympathetic blockade. No significant changes in man systemic B.P. and pulse rate were observed pre- and post-POB administration. Noradrenaline was

capable of producing a systemic pressor response in the POB pretreated group comparable to that seen in the non-POB treated group. Right renal vascular resistance was increased during noradrenaline infusion even in the POB pretreated kidney. A number of explanations can be offered. As mentioned previously, prolonged noradrenaline infusion can produce marked plasma volume depletion and shock (Sutter, 1963). All animals showed significant increases in central venous hematocrit, probably partly reflecting a decrease in circulating volume. Noradrenaline infusion can also be responsible for the release of other vasoactive substances, particularly renin, resulting in the generation of angiotensin II (Vander, 1965). Phenoxybenzamine is known to be ineffective in blocking the renal vascular effects of angiotensin II. This could, in part, explain the increase in renal vascular resistance during noradrenaline-induced hypertension. Since autoregulation is still present in the denervated kidney and the kidney treated with sympathetic blocking agents, the poorly-understood phenomenon of renal autoregulation could also be partly or entirely responsible.

The failure to induce functional acute renal failure in the kidney blocked with POB implies that it is the alpha-agonistic properties of noradrenaline, and likely their vascular effects following alpha receptor activation, that are responsible for the production of renal failure in this experimental model and it is unlikely to be due to direct (cellular)

TABLE XIII

Effect of Intra-arterial Noradrenaline Infusion on Renal Hemodynamics and Renal Function in the Dog Pretreated with Intra-arterial Phenoxybenzamine
N=4

Right Kidney	Control (0.5 hrs)	Post-POB (1.0 hrs)	NA Infusion (2.0 hrs)	2 Hrs. Post-NA (2.0 hrs)	4 Hrs. Post-NA (2.0 hrs)
RBF (% Control)	100	103.3 (3.3)	93.1 (5.0)	98.4 (11.1)	97.7 (7.9)
Mean B.P. (mm Hg)	130.0 (5.8)	135.0 (2.9)	151.7* (11.7)	128.3 (7.2)	123.3 (4.4)
RVR (% Control)	100	93.9 (6.1)	118.9* (17.3)	97.3 (16.8)	92.9 (13.5)
C_{Cr} (% Control)	100	107.5 (13.4)	92.0 (14.9)	85.0 (10.5)	119.0 (23.0)
$\frac{\dot{V} \text{ (Right)}}{\dot{V} \text{ (Left)}}$	1.24 (0.35)	1.30 (0.44)	1.03 (0.18)	1.31 (0.53)	1.19 (0.56)

Values expressed as Mean (S.E.)
Paired-t test used for statistical analysis.
* $P < 0.05$
Values without p Values attached = No significant difference from control.

TABLE XIV

Renal Hemodynamics and Renal Function
Following Intra-arterial nor Noradrenaline Infusion:
Phenoxybenzamine Pretreatment Versus Control

Right Kidney	Control			NA Infusion			4 Hrs. Post-NA		
	Control	POB	p	Control	POB	p	Control	POB	p
RBF (% Control)	-	-	-	1.9 (1.4)	93.1 (5.0)	<0.001	70.2 (4.8)	97.7 (7.9)	<0.02
Mean B.P. (mm Hg)	129.4 (4.7)	140.0 (5.8)	NS	168.9 (9.1)	151.7 (11.7)	NS	122.2 (4.5)	123.3 (4.4)	NS
RVR (% Control)	-	-	-	-	118.9 (17.3)	<0.001	140.4 (15.0)	92.9 (13.5)	NS
C_{Cr} (% Control)	-	-	-	0 (0)	92.0 (14.9)	<0.001	14.7 (11.7)	119.0 (23.0)	<0.001
$\frac{\dot{V} \text{ (Right)}}{\dot{V} \text{ (Left)}}$	0.84 (0.18)	1.24 (0.35)	NS	0.02 (0.12)	1.03 (0.18)	<0.001	0.88 (0.43)	1.19 (0.56)	NS

Values expressed as Mean (S.E.)
Non-paired t-test was used for statistical analysis.
Control = non-POB pretreated dog, N=9.
POB = POB pretreatment, N=4.

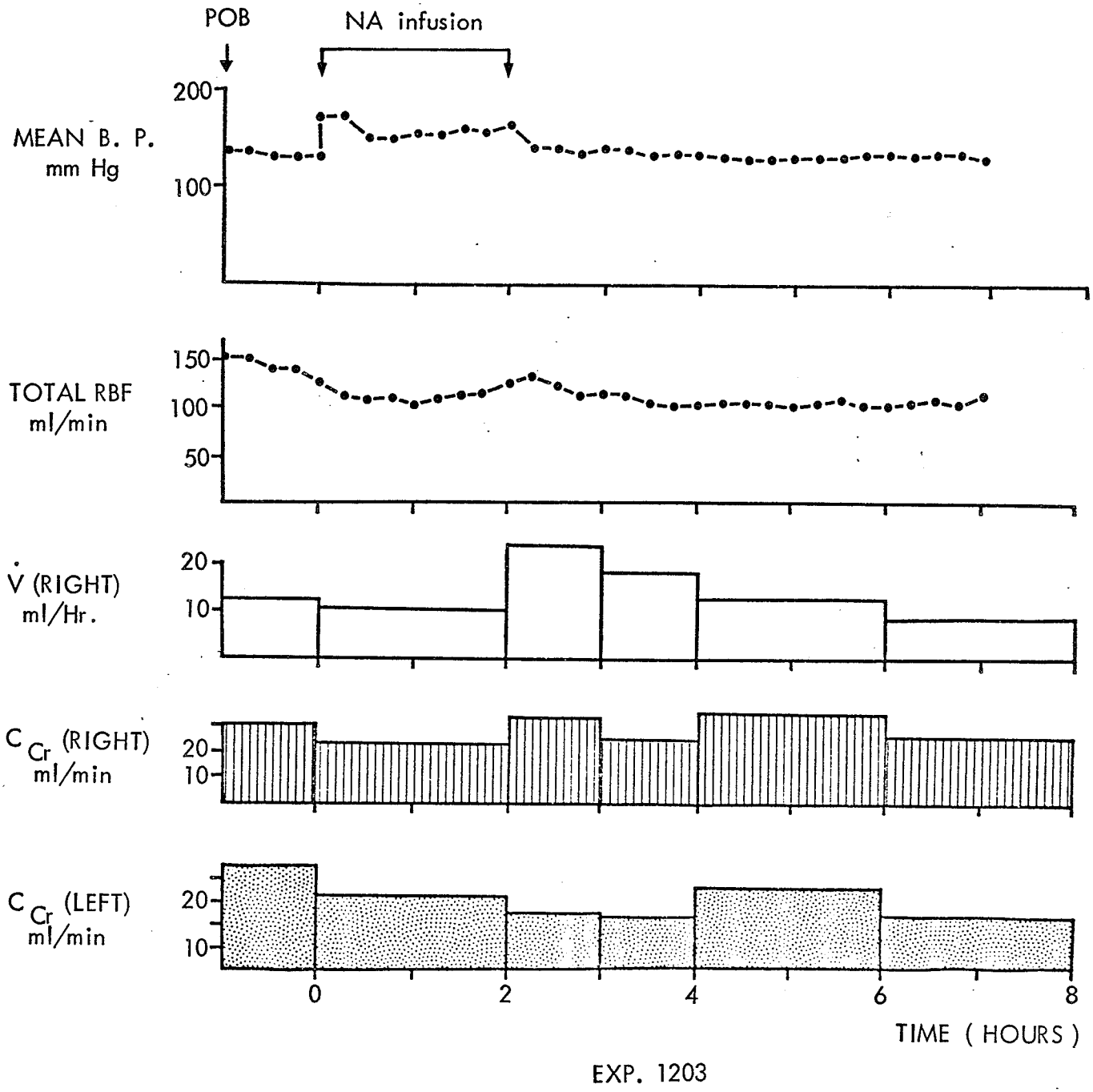


Figure 8: Renal hemodynamics and renal function following intra-arterial noradrenaline infusion in the dog pre-treated with intra-arterial phenoxybenzamine.

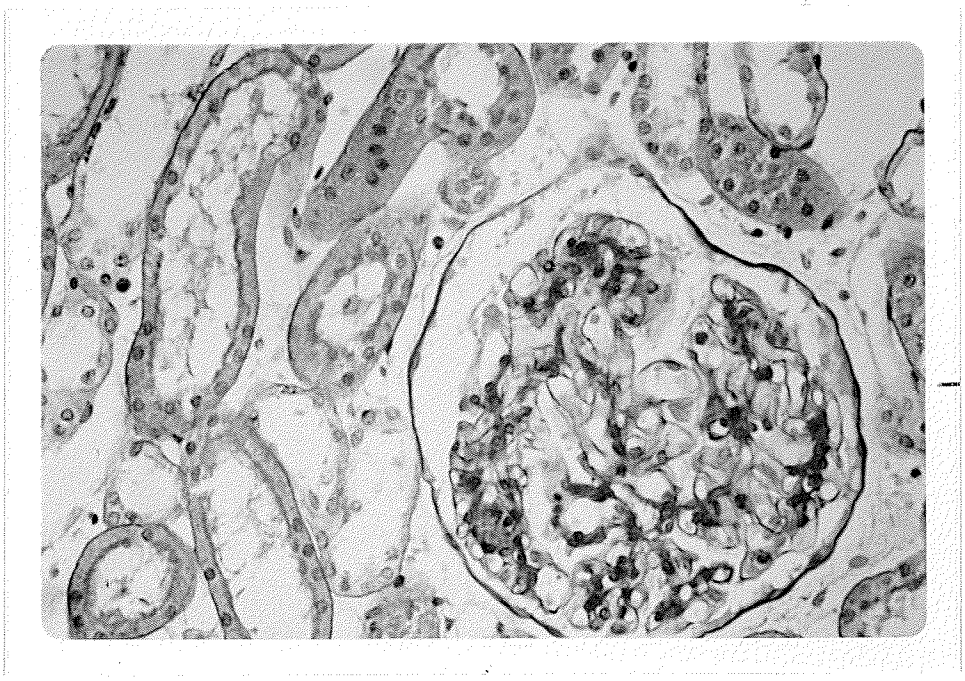


Figure 9: Histological appearance of right kidney pre-treated with phenoxybenzamine following intra-arterial noradrenaline infusion (Exp. 1202). Tubular necrosis and debris in tubular lumen can be seen. Glomerulus appear normal. Glomerular filtration was well-maintained throughout.

effects of noradrenaline per se. The presence of histologic tubular damage in the POB pretreated kidney supports the hypothesis that the primary lesion in acute renal failure is a reduction in glomerular filtration rather than due to structural tubular damage.

5. Effects of Intra-arterial Infusion of Angiotensin II

The Renin-Angiotensin axis has been implicated in the pathogenesis of acute oliguric renal failure by numerous authors (Sevitt, 1959; Schnerman et al. 1966; and Brown et al. 1970). Plasma renin activity has been found to be elevated in clinical acute renal failure (Tu, 1965; Kohot and Kuska, 1969; and Brown et al. 1970). Angiotensin has been reported to cause renal vascular damage (Byrom, 1964). Furthermore, studies in glycerol-induced acute renal failure in the rat have shown that chronic salt loading with suppression of renin production resulted in a less severe lesion (Thiel et al. 1970; McDonald et al. 1970; and DiBona et al. 1971). Since noradrenaline infusions are known to result in elevated plasma renin-activity (Vander, 1965), we have attempted to determine whether prolonged intra-arterial infusions of angiotensin in large doses can produce acute renal failure as is in the case with noradrenaline.

a. Methods:

The general procedure was as outlined in Section III, 2 (i) with the following modifications: Three mongrel dogs of either sex, weighing 15-25 kg were used. Following stabilization and a 60 min control

period, angiotensin II amide (Hypertensin^{R}- CIBA) was infused directly into the right renal artery for 45 min by a Harvard infusion-withdrawal pump at a dose of at least 7.5 ug/min/kg B.Wt. This dose is capable of producing marked systemic hypertension initially, comparable to that seen in experiments with noradrenaline infusions. Urine and blood samples were collected during control, and infusion periods, and for 2 hours post-infusion.

b. Results:

Renal hemodynamic and renal function data are shown in Table XV. Intra-arterial infusion of angiotensin resulted in systemic hypertension with an initial decrease in total renal blood flow, followed by a subsequent but incomplete recovery for the rest of the infusion period. In all animals, renal blood flow was relatively well-maintained. Renal vascular resistance was moderately elevated during the infusion period. Renal function demonstrated only transient disturbances. A marked diuresis was observed with high rates of infusion of angiotensin. No functional or histological evidence of renal failure was observed at the end of the experimental period.

c. Comments:

Angiotensin is well known as a powerful vasoconstrictor. Carriere and Friborg (1969) have shown that intra-arterial administration of angiotensin resulted in a decrease in renal blood flow with

TABLE XV

Effect of Intra-arterial Angiotensin II on Renal Hemodynamics and Renal Function in the Dog
N=4

Right Kidney	Control Period	Angiotensin Infusion	2 Hrs. Post-infusion	4 Hrs. Post-infusion
RBF (% Control)	100	71.2* (9.9)	89.3 (7.7)	100.6 (7.7)
Mean B.P. (mm Hg)	130.0 (10.4)	163.3* (19.7)	120.0 (5.0)	116.7 (4.4)
RVR (% Control)	100	186.8* (31.0	104.9 (5.2)	90.4 (4.2)
$\dot{V}$ ml/hr (% Control)	100	258.0* (65.4	114.6 (28.6)	92.1 (36.1)
C_{Cr} ml/min (% Control)	100	65.5* (14.3)	83.0* (8.8)	86.0 (2.4)

Results expressed as Mean ± S.E.
NS = Not significant
* $P < 0.05$
Paired-t test was used for analysis.

cortical ischemia in the dog. However, the drug was only infused for 15 minutes in their study. Prolonged administration of angiotensin results in tachyphylaxis (Bock and Gross, 1961). In the present study, tachyphylaxis in the renal vascular bed appeared after 15 minutes and additional increases in rate of infusion failed to maintain initial reduction in renal blood flow. This is in agreement with the findings of Villareal et al. (1964). A diuresis with increase in sodium excretion was observed under our experimental conditions during angiotensin infusion. This is in accord with the findings of other investigators (reviewed by Bock et al. 1968).

It can be concluded that angiotensin, although present in high concentrations in the renal circuit, cannot be singularly responsible for the production of acute renal failure. However, this does not negate its possible synergistic role in the noradrenaline-induced lesion. This may be due to a difference in the susceptibility in the development of tachyphylaxis between the angiotensin receptor and the noradrenaline receptor.

6. Effect of Intra-arterial Infusion of Vasopressin

High levels of circulating vasopressin are present following hemorrhage, shock etc. (Ginsburg and Brown, 1956; Beleslin et al. 1967) where acute renal failure is likely to develop. Circulating vasopressin has been shown to be responsible for a considerable portion of the

splanchnic vasoconstriction in the cat in hemorrhagic hypotension (Greenway and Stark, 1969; McNeill et al. 1970). Byrom (1937) reported histological damage to rat kidneys following repeated intraperitoneal administration of pitressin. Although its effects on renal water handling are well known, the effects of vasopressin on the renal circulation are still obscure. Aukland (1968a) reported no change in renal perfusion or re-distribution of intrarenal blood flow following intravenous administration of vasopressin in the dog. However, Wakim et al. (1942) have reported a decrease in renal blood flow. Intrarenal blood flow redistribution has also been reported by Fischer et al. (1970) following intra-arterial administration of vasopressin. We have attempted to determine whether intra-arterial administration of vasopressin would result in renal ischemia of an order of magnitude similar to that produced by noradrenaline, and whether it was capable of producing renal failure with prolonged infusion.

a. Methods:

The general procedure was as outlined in Section III, B with the following modifications: Four mongrel dogs of either sex weighing 15-20 kg were used. Following stabilization, and a 60 minute control period, vasopressin (Pitressin^{R}- Parke, Davis) at a rate of 5.0 uUnits/min/kg, was administered by a constant infusion-withdrawal pump into the right renal artery. This rate of infusion was sufficient to produce

a systemic pressor effect and cardiac arrhythmias which consisted of prolonged P-R intervals and supraventricular extrasystoles. Infusion lasted 45 minutes. Urine and blood samples were collected during the control period, during infusion and for 4 hours post-infusion.

b. Results:

Despite increases in systemic arterial blood pressures, renal blood flow showed little change during the infusion period (Table XVI). Calculated renal vascular resistance was increased. No significant changes in glomerular filtration rate were observed during and following infusion. Urine flow rate increased markedly from both kidneys during and following infusion of vasopressin. No evidence of renal failure was observed 2 hours post-infusion.

c. Comments:

Although the renal vascular effects of vasopressin are not well-known, it is apparent that I. A. administration of vasopressin in the anesthetized dog did not produce drastic changes in renal blood flow or function of the order of magnitude as that observed with noradrenaline. The diuretic effects of ADH in the anesthetized animals have previously been reported (Brooks and Pickford, 1958; McDonald and de Wardener, 1965; and Martinez-Maldonado et al. 1971). The diuresis observed from the left kidney during infusion of vasopressin into the right renal artery was due to the systemic effects of vaso-

TABLE XVI

Effect of Intra-arterial Vasopressin on Renal Hemodynamics and Renal Function in the Dog
N=4

Right Kidney	Control Period	Vasopressin Infusion	2 Hrs. Post-infusion
RBF (% Control)	100	97.7 (2.3)	107.3 (6.0)
Mean B.P. (mm Hg)	130.0 (5.2)	146.7* (8.8)	125.0 (5.8)
RVR (% Control)	100	115.5 (5.0)	89.9 (2.3)
$\dot{V}$ ml/hr (% Control)	100	133.2* (51.1)	304.8* (219.6)
C_{Cr} ml/min (% Control)	100	109.4 (1.8)	111.3 (11.3)

Results expressed as Mean $\pm$ S.E.
NS = Not significant
* $P < 0.05$

pressin in the anesthetized dog. With 45 minutes of vasopressin infusion, considerable quantity of drug was present in the animal. This explains the diuresis even after infusion of vasopressin had stopped. The dosage of vasopressin used was high enough to produce systemic pressor and cardiac effects and probably represents the ceiling dose possible in the whole animal. It can be concluded that vasopressin cannot be singularly responsible for the production of acute renal failure in the dog.

7. Discussion

The foregoing studies have demonstrated that a 2 hour period noradrenaline infusion, at a dosage sufficient to reduce total renal blood flow to near zero during the period of infusion, results in the production of a unilateral renal lesion showing the morphologic, hemodynamic and functional characteristics of human acute renal failure. This model was shown to be readily reproducible. Noradrenaline presumably induces its effects via the vascular alpha-adrenergic receptors in the renal circuit resulting in a sustained high resistance to blood flow and disporportionate reduction in glomerular filtration.

In equi-pressor doses, acute renal failure could not be produced by intravenous infusion of noradrenaline, or the intrarenal arterial infusion of angiotensin II or vasopressin.

C. The Role of Renin-angiotensin in the Development of Acute Renal Failure

Although the pathogenesis of oliguria in acute renal failure is still poorly understood, numerous lines of evidence have implicated a reduction in renal blood flow and glomerular filtration as being the primary events (Flanigan and Oken, 1965; Ruiz-Guinazu et al. 1967; Hollenberg et al. 1968; and DiBona et al. 1971). It was first postulated by Goormaghtigh (1945, 1947) that increased concentration of angiotensin formed in the region of the glomerulus might reduce renal blood flow, thereby precipitating acute renal failure. This theory has been further supported by the work of Schnermann and Coworkers (1966). Furthermore, chronic saline loading, presumably associated with renin suppression has been shown to be partially protective in glycerol induced (McDonald et al. 1970; Thiel et al. 1970) and mercuric chloride induced (DiBona et al. 1970) acute renal failure in the rat. Previously, we have demonstrated that prior renal denervation and post-event phenoxybenzamine administration were partially protective in the glycerol-rat model. Since sympathetic discharge causes renin release (Vander, 1965; Bunag et al. 1966b) an attempt was made to determine the effect of chronic saline loading on the subsequent development of the lesion and the combined effect of chronic saline loading and prior denervation.

1. Glycerol-induced Acute Renal Failure in the Rat.

(i) Effect of Chronic Saline Loading

a. Methods:

The general experimental protocol was as outlined in Section III, A with the following modifications: Nine female Long Evans rats, weighing approximately 300 gms were given 0.9% saline (ad libitum) in place of drinking water for at least 4 weeks prior to experimentation. Three days prior to dehydration (being part of the preparation for glycerol injection), tap water for drinking was resumed. Renal function from the period 24 to 48 hours post-glycerol administration was compared to a water-drinking control group subjected to the same dose of glycerol (28% Glycerol, at 10 mg/kg).

b. Results:

Renal function for the period 24 to 48 hours post-glycerol administration for the saline-drinking group and the water-drinking controls are shown in Table XVII. Body weights of both groups were comparable. In the saline loaded animals, mean creatinine clearance was higher, mean plasma creatinine concentration lower, and free water production and % TRF_{Na} were lower than the control group. However, these differences were not statistically significant.

c. Comments:

It has been shown that chronically increased dietary salt content resulted in suppression of both renin synthesis and release

TABLE XVII

Effect of Chronic Saline Loading on Glycerol-induced Acute Renal Failure in the Rat. Renal Function at 24 to 48 Hours Post-glycerol Administration

	Control (n=20)	Saline Drinking (n=9)	p Value
$\dot{V}$ (ml/100 gm/24 hrs.)	9.7 ± 1.3	7.7 ± 1.7	NS
C_{Cr} (ml/100 gm/24 hrs.)	60.9 ± 21.6	84.8 ± 39.0	NS
C_{osm} (ml/100 gm/24 hrs.)	9.4 ± 1.3	8.0 ± 1.8	NS
C_{H_2O} (ml/100 gm/24 hrs.)	13.8 ± 2.4	10.6 ± 3.5	NS
U_{osm} (mOsm/kg)	426.1 ± 22.5	420.5 ± 41.6	NS
% TRF_{Na} (%)	39.5 ± 11.5	28.9 ± 8.4	NS
P_{Cr} (mg%)	5.65 ± 0.65	4.78 ± 8.78	NS

Control = water drinking rats (28% glycerol, 10 ml/kg).

NS = Not Significant at 5% level.

(Vander, 1965; Gross et al. 1966b). Also, renal renin depletion has been reported with chronic salt loading (Gross et al. 1964). On the basis of these findings, it has been suggested that the reduction or absence of renin-angiotensin activity accounted for the "protective effect" of salt loading on the development of acute renal failure in the glycerol treated rat. It must be noted, however, that the effects of chronic salt loading are many. We have found that the degree of renin depletion is variable and incomplete. Animals given 0.9% saline as drinking water tend not to drink as much for obvious reasons. This lead to some degree of plasma volume reduction and hemoconcentration. Mean hematocrit of 10 rats after 4 weeks of 0.9% saline drinking was 56% compared to a mean hematocrit of 40% in water drinking animals. Because of this, tap water for drinking was resumed for 3 days prior to experimentation. This maneuver resulted in a return of hematocrit to normal, but probably a lesser degree of renin depletion was also achieved.

Chronic salt loading, associated with mineralocorticoid function, may produce systemic hypertension (Gross et al. 1965; Page and McCubbin, 1968; and Pickering, 1968) and changes in vascular smooth muscle reactivity (Friedman and Friedman, 1963). Less well-defined relationships exist between increased salt intake and intrarenal blood flow distribution (Earley and Friedler, 1965; Horster and Thurau, 1968;

Abbrecht et al. 1969 and Mogil et al. 1969). Hollenberg et al. (1970b, 1971) reported an increase in renal cortical blood flow in normal subjects on high salt diets. This pattern of intrarenal blood flow distribution was also seen in some hypertensive individuals with normal renal function even on salt restricted diets (Hollenberg and Merrill, 1970). It is quite conceivable that all these factors may influence the outcome of glycerol-induced acute renal failure, perhaps in an adverse manner, counteracting the beneficial effects of renin-depletion. Chronic saline loading may be a poor way to study the effects of renin depletion on acute renal failure in this model.

In summary, we have been unable to show any significant effect of prior saline loading alone on the changes in renal function induced by I.M. glycerol in the rat. One might suspect, however, that a slight protective effect is present. This effect may be too small for the experimental method employed to detect.

(ii) Effect of Chronic Saline Loading and Prior Unilateral Renal Denervation

Either renal denervation (Ueda et al. 1967) or chronic salt loading (Gross et al. 1964) has been shown to cause a reduction of renal renin content. In addition, numerous studies have demonstrated directly or indirectly that a relationship exists between sodium homeostasis, the sympathetic nervous system and renal function (Kamm and Levinsky, 1965; Gill and Bartter, 1966; Schrier et al. 1967; Romero et al. 1968; Bonjour et al. 1969; Hollenberg et al. 1970b, 1971; and

Lackner and McKay, 1971). It is tempting to suggest that an interaction exists between the sympathetic nervous system and the renin-angiotensin system in the pathogenesis of acute renal failure. In an attempt to investigate this possibility, the combined effects of chronic salt loading and prior renal denervation on glycerol induced acute renal failure in the rat were studied.

a. Methods:

The general experimental procedure was as outlined in Section III, A with the following modifications: Ten female Long Evans rats, weighing approximately 300 gms were used. Animals were given 0.9% saline as drinking water ad libitum for 3 weeks prior to denervation of the left kidney. At least 1 week was allowed for recovery from surgery. Tap-water for drinking was resumed 3 days prior to dehydration and subsequent glycerol injection. Renal function 24 to 48 hours post-glycerol administration was compared to a group of innervated water-drinking controls given the same dosage of glycerol (28% glycerol, 10 ml/kg B.Wt.).

b. Results:

Table XVIII shows the renal functional parameters in the unilaterally denervated saline-drinking group and the innervated water-drinking controls 24 to 48 hours post-glycerol administration.

Mean endogenous creatinine clearance, osmolar clearance,

urine osmolality show significantly higher values in the treated group. Free water production, % TRF_{Na} and plasma creatinine concentration are significantly lower. These findings indicate less severe derangement in function in the unilaterally denervated, saline loaded group as compared to the unmodified control. In addition, when the unilaterally denervated saline loaded group was compared to the saline drinking group, better preservation of renal function in the former group was observed.

c. Comments:

It is evident that while chronic saline loading alone was not associated with significant protection against acute renal failure, it may potentiate the protective effect of denervation. Whether the renin angiotensin system is necessarily involved is unclear.

TABLE XVIII

Effect of Chronic Saline Loading and Prior Unilateral Renal Denervation on Glycerol-induced Acute Renal Failure. Renal Function at 24 to 48 Hours Post-glycerol Administration

	Control (n=20)	Saline Drinking and Unilat. Denervation (n=9)	p Value
$\dot{V}$ (ml/100 gm/24 hrs.)	9.7 ± 1.3	6.3 ± 1.3	NS
C_{Cr} (ml/100 gm/24 hrs.)	60.9 ± 21.6	515.7 ± 93.5	< 0.001
C_{osm} (ml/100 gm/24 hrs.)	9.4 ± 1.3	17.2 ± 1.3	< 0.001
C_{H_2O} (ml/100 gm/24 hrs.)	13.8 ± 2.4	-1.93 ± 2.0	< 0.001
U_{osm} (mOsm/kg)	426.1 ± 22.5	1029 ± 154	< 0.001
% TRF_{Na} (%)	39.5 ± 11.5	1.7 ± 0.4	< 0.001
P_{Cr} (mg%)	5.65 ± 0.65	0.85 ± 0.25	< 0.001

Control = water drinking rats (28% glycerol, 10 ml/kg).

NS = Not Significant at 5% level.

(iii) General Comments:

In the rat model, it has been demonstrated by a number of investigators that numerous factors such as state of hydration (Thiel et al. 1966, 1967), osmotic diuresis (Teschan and Lawson, 1966; Wilson et al. 1967, 1969), and state of sodium homeostasis (McDonald et al. 1970; DiBona et al. 1971), can favorably influence the severity of acute renal failure. All of these "protective" manipulations necessitated gross physiological readjustment. This makes the mechanism of protection obscure. We have shown previously that prior renal denervation or early post-event phenoxybenzamine administration partially protected against acute renal failure. This implies that the sympathetic nervous system plays a role in the production of the lesion. All of the factors mentioned above shown to be protective involve reflex suppression of sympathetic overactivity either directly or indirectly. McGiff and Fasy (1965) have reported that denervation abolished the renal vasoconstriction produced by intravenous injection of angiotensin. While a multifactorial situation is generally involved in clinical acute renal failure, it is conceivable that a final common pathway may involve the activation of the adrenergic nervous system as a significant contribution in the rate limiting process.

This is further demonstrated by the synergistic protective effects of chronic saline loading and prior renal denervation. A

graphical summary of the pathogenetic factors studied in the glycerol-rat model is shown in Figure 10. Plasma creatinine concentration and endogenous creatinine clearance are chosen as parameters of glomerular function, and osmolar clearance and % TRF_{Na} are chosen to be indices of tubular impairment. It is apparent that, under the given experimental conditions, a trend of increasing effectiveness in protecting against acute renal failure can be observed. The fact that prior denervation with saline loading is more effective than denervation alone further supports the impression that saline-loading itself may be a biological significant manipulation.

One must safeguard against the assumption of equating chronic saline loading with renin-depletion. Protection experiments of these types cannot establish a definitive pathogenetic role of renin-angiotensin in acute renal failure.

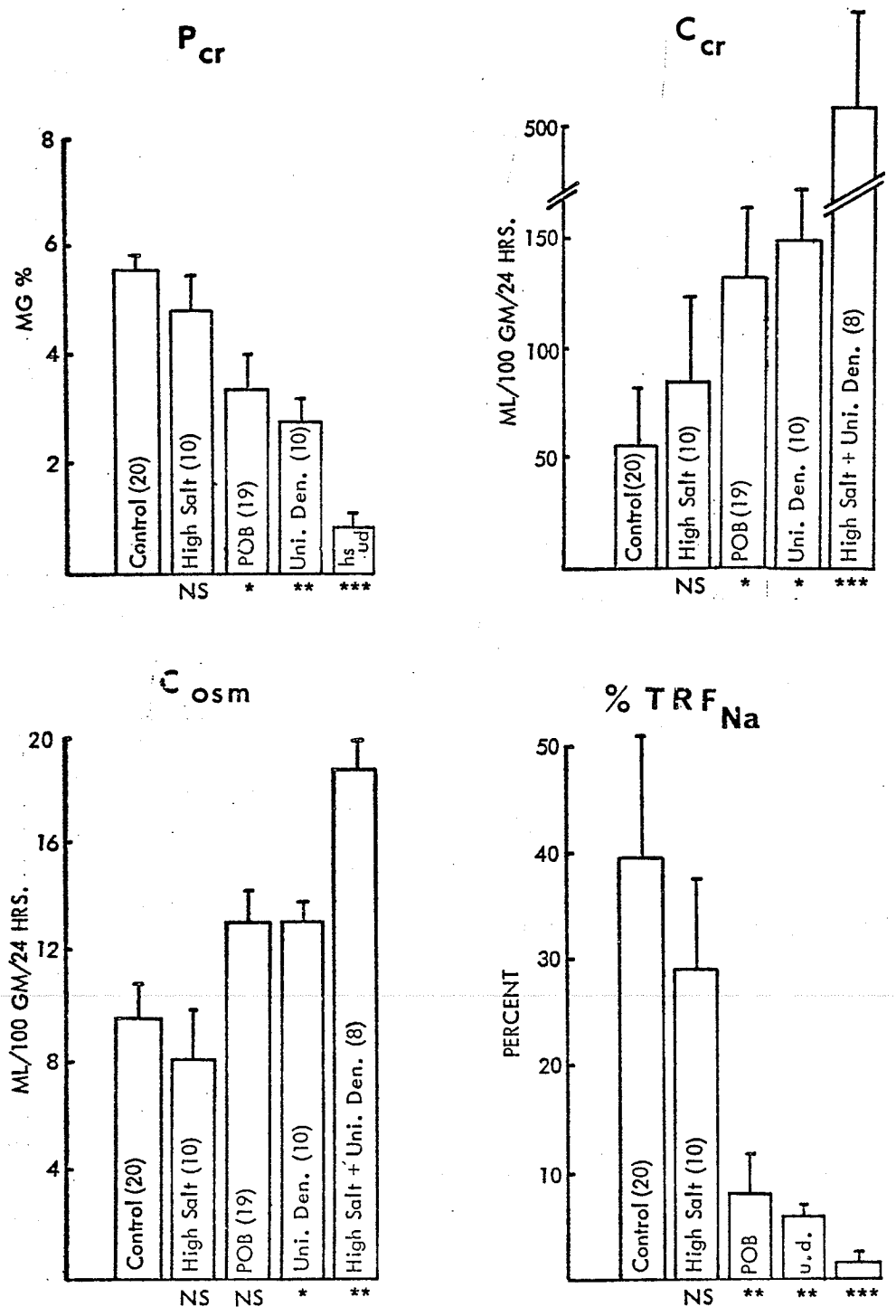


Figure 10: Glycerol-induced acute renal failure in the rat. Histograms represent means ± S.E.; number of rats in each group in parenthesis.
Control = Unmodified group, I.M. 28% glycerol, 10 ml/kg B.Wt.
Hi Salt = Chronic 0.9% saline drinking group before glycerol injection.
POB = I.P. POB given 2.5 hours post-glycerol injection.
Uni. Den. = Prior unilateral denervation.
* $P < 0.05$ ** $P < 0.01$ *** $P < 0.005$
NS = Not Significant at 5% level.

2. Noradrenaline-induced Acute Renal Failure in the Dog.

Despite the popularity of the renin-angiotensin theory in the pathogenesis of acute renal failure (Sevitt, 1959; Schnermann et al. 1966 and Brown et al. 1970), there is little direct evidence to substantiate this claim. Little angiotensin II is generated through one single passage of the renal circulation (Ng and Vane, 1968). Prolonged intravenous infusions of angiotensin II do not produce acute tubular necrosis (Sutter, 1963). In an attempt to evaluate the role of the renin-angiotensin axis in the production of acute renal failure, in the noradrenaline-dog model, plasma renin activity was determined under several different experimental conditions.

(i) Effect of Prolonged Infusions of I. V. Noradrenaline, I. A. Noradrenaline, and I. A. Vasopressin on Plasma Renin Activity

a. Methods:

Plasma samples for the determination of renin activity (PRA) were obtained from animals randomly selected from three groups of experiments detailed in Section III, B (during infusions of I. V. noradrenaline, I. A. noradrenaline and I. A. vasopressin). Samples were obtained during control periods, during infusions of test drug and at 2 and 4 hours thereafter. For comparison, blood samples were also obtained from 7 healthy conscious mongrel dogs fed a similar diet. Samples in these latter animals were obtained following 14 hours of fasting, but with free access to drinking water.

b. Results:

Table XIX shows the values for plasma renin activity (ng angiotensin II/ml/min) in three groups of animals. It can be seen that plasma renin activity is markedly elevated during intra-arterial infusion of noradrenaline and for the remainder of the post-infusion period following the development of unilateral acute renal failure. Intravenous infusions of noradrenaline at the same dosages showed comparable levels of increases in plasma renin activity. These animals did not develop acute renal failure. No significant change in plasma renin activity was observed during and after intra-arterial infusions of vasopressin.

c. Comments:

It can be assumed that when angiotensin was infused into the renal artery at a rate sufficient to raise systemic blood pressure it was probably also present in high concentrations in the renal circuit. This, and the previous observation that renal failure did not develop with intra-arterial angiotensin infusion do not lend support to the theory that the renin-angiotensin system is significantly responsible for the production of acute renal failure in this experimental model. While prolonged intravenous noradrenaline results in similar magnitudes of increases in P.R.A. as those in intrarenal noradrenaline, it can be concluded from our results that marked elevation of plasma renin

TABLE XIX

Effect of Intravenous Noradrenaline, Intra-arterial Noradrenaline and Intra-arterial Vasopressin on Plasma Renin Activity in the Dog

	Control	During Infusion	2 Hrs. Post-infusion	4 Hrs. Post-infusion
Group I				
I. A. Noradrenaline (n=8)	8.9 (1.5)	36.3* (13.8)	47.1** (9.7)	26.9* (9.5)
Group II				
I. V. Noradrenaline (n=3)	8.3 (0.2)	24.8** (2.7)	24.9* (9.9)	16.2 (3.3)
Group III				
I. A. Vasopressin (n=3)	7.1 (2.0)	6.4 (0.1)	7.2 (1.5)	11.5 (2.4)
Normal Values (n=7)		13.2 (3.6)		

Results expressed as Mean (S. E.)
Paired-t test was used for statistical analysis within the same group.
* $P < 0.05$ ** $P < 0.01$
Values without p Values attached = No significant difference from control period.
(Non-paired t test was used for statistical analysis between groups. Statistical differences are presented in text.)
P. R. A. - measured in ng Angiotensin I generated/ml/min.

activity is not necessarily associated with the production of acute renal failure. Vasopressin has been shown to be effective in inhibiting renin release (Bunag et al. 1967; Tagawa et al. 1971). This explains the lack of increase in plasma renin activity during and following infusions of vasopressin in the above experiment.

(ii) Effect of Short-term Dietary Salt Supplement on Renal Hemodynamics and Plasma Renin Activity in Noradrenaline-induced Acute Renal Failure

Chronic saline loading, associated with renin depletion, offers partial protection in glycerol-induced acute renal failure (Henry et al. 1968; McDonald et al. 1969; Thiel et al. 1970; DiBona et al. 1971). In an attempt to delineate the individual effects of salt loading and renin depletion on the outcome of acute renal failure, the effect of intrarenal artery infusion of noradrenaline on renal hemodynamics and renal function was studied in 5 mongrel dogs on short-term dietary salt supplement.

a. Methods:

The general experimental procedure was as outlined in Section III, B with the following modifications: Five mongrel dogs of either sex, weighing 15-26 kg were used. Dietary salt supplement in the form of NaCl tablets approximately 0.1 gms/day/kg body weight were given for 7 days prior to experimentation. Renal hemodynamics, renal function and plasma renin activity were compared to a control group

fed with standard laboratory chow pellets only.

b. Results:

Results are shown in Table XX. Infusions of noradrenaline directly into the renal artery always resulted in complete renal ischemia and anuria during infusion in the standard model. In contrast, animals who had received prior dietary salt supplementation maintained considerable renal blood flow. Complete ischemia could not be achieved even with further increase in the rate of noradrenaline infusion. Rates as high as 970 ug/min were used. Higher rates of infusion were prevented by the development of marked hypertension (mean B. P. up to 250 mm Hg) and cardiac arrhythmias (nodal tachycardia, bigeminal rhythm and multifocal ventricular premature contractions). Despite considerable increase in renal vascular resistance, urine output continued during the entire infusion period.

In addition, during the post-noradrenaline infusion period, renal blood flow and creatinine clearance were higher in those animals who received salt supplement than in control animals. The sequence of hemodynamic and functional events of a typical experiment is shown in Figure 11.

Plasma renin activity in the two groups of animals did not differ significantly during control period, infusion of noradrenaline or during post-infusion.

TABLE XX

Renal Hemodynamics, Renal Function and Plasma Renin Activity
Following Intra-arterial Noradrenaline Infusion in the Dog:
Dietary Salt Supplement Versus Control

Right Kidney	Control Period			NA Infusion			2 Hrs. Post-NA			6 Hrs. Post-NA		
	Control	Salt	p	Control	Salt	p	Control	Salt	p	Control	Salt	p
RBF (% Control)	100	100	-	2.0 (1.4)	22.8 (15.1)	<0.05	53.9 (8.3)	53.4 (7.4)	NS	69.6 (10.0)	72.0 (20.6)	NS
Mean B.P. (mm Hg)	129.4 (4.7)	128.8 (8.3)	NS	168.9 (9.1)	158.8 (8.3)	NS	126.7 (6.3)	130.0 (7.4)	NS	119.4 (5.4)	122.5 (22.5)	NS
RVR (% Control)	100	100	-	-	-	-	217.5 (40.0)	222 (45.8)	NS	131.7 (10.9)	148.7 (36.6)	NS
$\dot{V}$ (% Control)	100	100	-	2.27 (2.12)	19.5 (4.9)	<0.01	21.1 (13.0)	22.4 (8.7)	NS	37.3 (15.0)	19.2 (6.6)	NS
C_{Cr} (% Control)	100	100	-	0	18.4 (7.9)	<0.01	3.6 (1.9)	50.6 (25.3)	<0.05	(12.7) (8.2)	47.0 (8.5)	<0.02
P.R.A. (ng/ml/min)	8.9 (1.5)	15.0 (4.8)	NS	36.3 (13.8)	64.2 (13.3)	NS	47.1 (9.7)	69.8 (14.2)	NS	26.9 (9.5)	33.9 (10.8)	NS
	N=8	N=4		N=8	N=4		N=8	N=4		N=8	N=4	

Control = Normal diet (N=9)
Salt = Dietary salt supplement 0.1 gm NaCl/day/kg (N=5)
Results expressed as Mean (S.E.).
Non-paired t test used for statistical analysis.
Plasma Renin Activity (P.R.A.) in 7 normal dogs = 13.18 $\pm$ 3.56 ng Angiotensin I/ml/min.

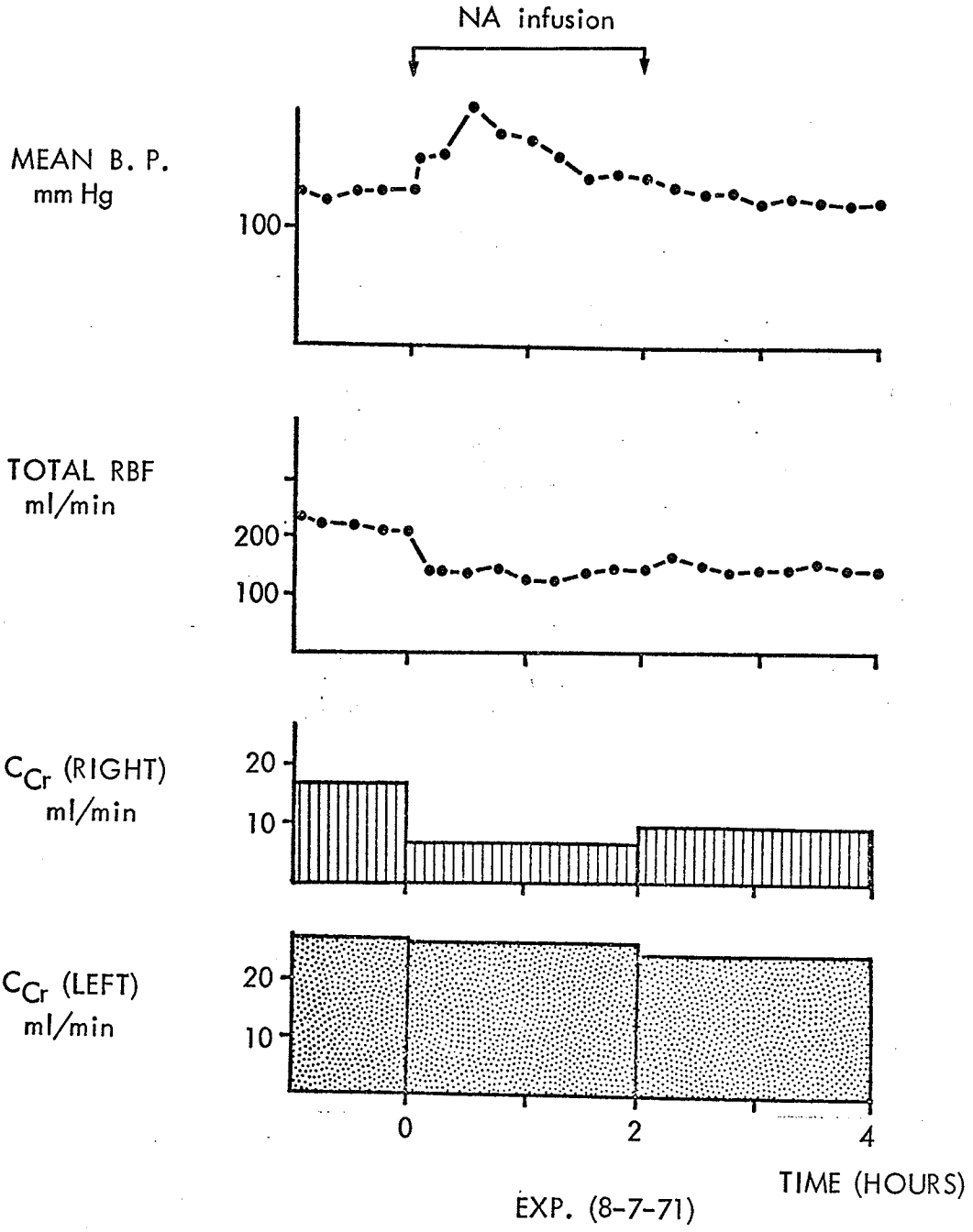


Figure 11: Renal hemodynamics and renal function following intra-arterial noradrenaline infusion in the dog pretreated with dietary salt supplementation.

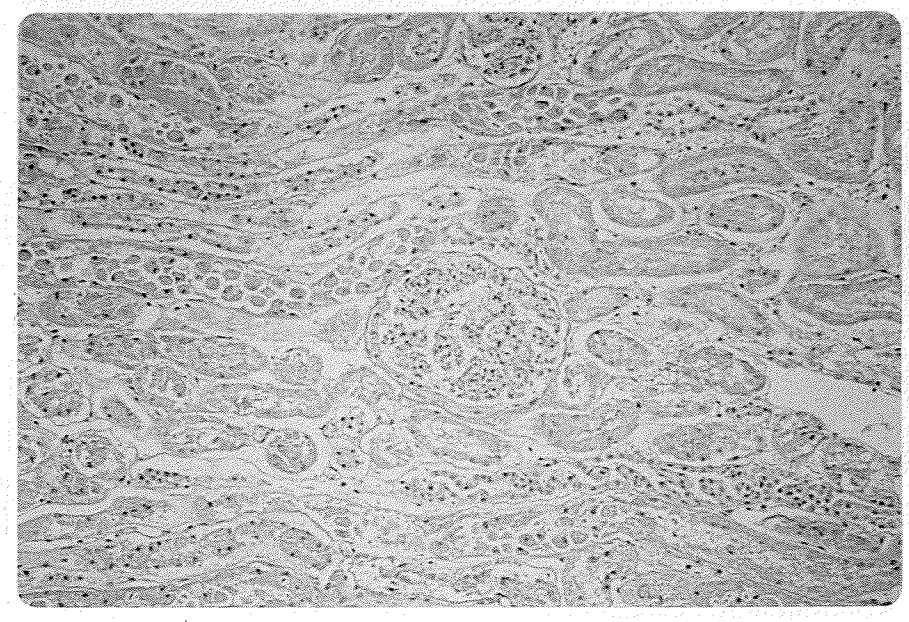


Figure 12: Histological appearance of right kidney 6 hours post-noradrenaline infusion in animal on dietary salt supplement (Exp. 15-7-71).
Marked intraluminal cellular desquamation can be seen. Glomerulus appears normal.

c. Comments:

Dietary sodium has been shown to modify renin release by noradrenaline (Bunag et al. 1966). Short-term dietary salt supplementation in the above experiment did not significantly suppress the increase in plasma renin activity following the infusion of noradrenaline. P.R.A. was markedly elevated in both the pretreated group and the controls. However, a remarkable qualitative difference in hemodynamic response to noradrenaline challenge was observed. The absence of complete ischemia in the salt supplemented group probably explains the lack of development of anuria and better post-event renal function.

Even with high intrarenal concentrations of noradrenaline and markedly elevated plasma renin activity, a difference in vascular responsiveness to the same insult was observed in the two groups of animals. This altered responsiveness was apparently sufficiently potent to resist the otherwise over-riding vascular insult of noradrenaline infusion. Thus, although the mechanism of the protective effect of salt-loading in unclear, it does not necessarily depend on its renin-depleting effects.

3. Discussion

The renin-angiotensin system has been proposed as being the mechanism responsible for the abnormal increase in renal vascular resistance in acute renal failure. However, much of the evidence used to support this claim is indirect.

Plasma renin activity and plasma angiotensin levels have been reported to be elevated in acute renal failure in man (Tu, 1965; Massani et al. 1966; Kohot and Kuska, 1969; Brown et al. 1970). Hemorrhage, hypotension, shock, bacterial toxins, surgical stress and renal artery occlusion have been shown to be potent stimuli for the release of renin (Vander, 1965; Brown et al. 1966; McKenzie et al. 1967; Vaughn et al. 1967). These are also situations where acute renal failure is most likely to occur (Merrill 1960, 1971). Goormaghtigh was the first to propose that an increase in angiotensin might reduce renal blood flow, thereby precipitating acute renal failure (Goormaghtigh 1947). Other authors (Sevitt, 1959; Finckh,1962; Schnermann et al. 1966; Henry et al. 1968 and Brown et al. 1970) have also proposed a similar role for the renin-angiotensin system. All of these theories represented variations of the same theme. The central issue involved a positive feedback mechanism where the RBF-lowering and GFR-reducing effects of angiotensin resulted in further release of renin. This postulate was further supported by the finding that the superficial cortex contained

the highest concentration of renin, less so in the juxtamedullary cortex, and absent in the medulla and the aglomerular subcapsular cortex (Cook, 1957; Cook and Pickering, 1959). This distribution of renin is similar to the distribution of structural damage in acute tubular necrosis (Brown et al. 1965). Also, chronic salt loading, associated with renin depletion offers partial protection in experimental acute renal failure (Henry et al. 1968; McDonald et al. 1969; Thiel et al. 1970; DiBona et al. 1971). However, several observations do not support the proposal that renin-angiotensin plays a significant role in the pathogenesis and maintenance of acute renal failure. Plasma renin activity is often, but not always elevated in patients with acute renal failure (Brown et al. 1970; Kohot and Kuska, 1969; McKenzie, 1971). Similar degree of increases are seen in situations such as malignant hypertension, which are not associated with the development of acute renal failure (Tu, 1965). Intravenous infusions of angiotensin even for prolonged periods of up to 2 hours do not result in acute renal failure (Nickerson and Sutter, 1964). It has been argued that plasma angiotensin concentrations tend to underestimate intrarenal concentrations of angiotensin (Brown et al. 1970). However, I. V. infusions of high doses of angiotensin results in the rapid development of tachyphalaxis in both man and the experimental animal (Day et al. 1965). Infusions of angiotensin in lower doses directly into the renal artery have re-

sulted in moderate reduction of renal blood flow without oliguria in the dog (Carriere and Friborg, 1969). With higher intra-arterial doses, we have been unable to produce acute renal failure due to the development of tachyphylaxis (Section III, experiment 5).

Although the mechanism of tachyphylaxis to angiotensin is poorly understood, the ease with which angiotensin induces tachyphylaxis is well known (Gross et al. 1968). It is difficult to conceive of a theory requiring on the one hand the presence of high local concentrations of angiotensin, and the absence of tachyphylaxis on the other. As well, the proposed positive feedback mechanism cannot explain the reversibility of the lesion. The relatively short transitional period between oliguria and diuresis in acute renal failure necessitates the sudden development of an escape phenomenon, usually occuring at a time when plasma renin activity is close to normal, or an alternative hypothesis.

We have demonstrated an altered renal response to prolonged intra-arterial infusions of noradrenaline in dogs pretreated with short-term dietary salt supplement. The finding that plasma renin activity was as markedly elevated as in dogs on normal diets during the experimental period would suggest a different protective mechanism other than renin depletion in acute renal failure and suggests that renin is not necessarily involved.

The interaction between noradrenaline and angiotensin is more complex. Liebau et al. (1966) showed that angiotensin tachyphylaxis in rat aortic strips could be reversed with the addition of noradrenaline in the bath. Strips from animals pretreated with reserpine showed rapid development of tachyphylaxis. However, no cross tachyphylaxis could be demonstrated between angiotensin and noradrenaline (Gross et al. 1961). This, together with our finding that in the rat, chronic saline drinking with renin depletion plus prior denervation offer more protection than denervation alone suggest that a possible significant interaction exists between the adrenergic nervous system and the renin-angiotensin system in the genesis of acute renal failure (Fung et al. 1970). Intravenous infusion of angiotensin has been reported to be capable of suppressing the release of renin secondary to potent stimuli such as hypotension, aortic and renal artery constriction (Vander et al. 1965; De Champlain et al. 1966). The existence of such a positive feed-back mechanism would mean that high plasma angiotensin levels are unable to suppress renin release, and therefore, a primary defect in the control of renin release would have to be postulated.

Thus, the unitarian theory as proposed by Brown et al. (1970) that renin-angiotensin is largely responsible for the initiation and the maintenance of acute renal failure cannot explain many important features of this lesion. More definitive progress will possibly be made with the development of an effective and specific angiotensin blocking agent.

D. Renal Vascular Responses in Acute Renal Failure.

The reduction in total renal blood flow to one third of normal in itself is not sufficient to explain the severely depressed renal function in acute renal failure. Comparable total renal blood flows occur in patients with chronic renal failure where the level of renal function is compatible with life (Hollenberg et al. 1968a). It has been demonstrated that maldistribution of intrarenal blood flow occurs in established acute renal failure (Hollenberg et al. 1968a, 1970), the hepatorenal syndrome (Epstein et al. 1970; Kew et al. 1971) and, renal allograft rejection (Lewis et al. 1967; Retik, 1967, 1968 and Rosen et al. 1968) where the degree of cortical ischemia was correlated with the decrease in renal function. Although the nature of this increase in renal cortical vascular resistance is still unknown, it is quite possible that different mechanisms are responsible for each of the different conditions. The presence of abnormal mediators has been postulated as an explanation in acute renal failure (Hollenberg et al. 1968). However, attempts to identify the mediators responsible have generally been met with little success.

Vasoactive agents, particularly adrenaline and noradrenaline can produce profound cortical ischemia (Block et al. 1952; Moses, 1952; Carriere, 1969; and Epstein et al. 1970b). Clinical evidence of enhanced adrenergic activity is frequently seen with the development of clinical acute renal failure. In addition, our findings suggest that the sym-

pathetic nervous system may be important in the pathogenesis of the lesion (Section III, A). Therefore, attempts were made to determine the effect of direct infusion of phenoxybenzamine into the renal artery on renal hemodynamics in established acute renal failure both in the dog and in man. As well, the renal vascular responses to intrarenal acetylcholine and acute volume expansion were also studied. The patterns of vascular responses were compared to that seen in the normal kidney. These studies may help to clarify the nature of some aspects of the disturbances in blood flow encountered in acute renal failure.

1. Noradrenaline-induced Acute Renal Failure in the Dog.

(i) Renal Vascular Responses to Acetylcholine

Acetylcholine when infused intra-arterially into the renal artery has been shown to produce dose-related increases in total renal blood flow in the normal dog (Vander, 1964; Pilkington et al. 1965; McGiff and Burns, 1967; Willis, 1968; Aukland and Logning, 1970; Carriere et al. 1971 and DiSalvo and Fell, 1971) and in normal man (Rashid et al. 1970; Freed et al. 1968). The dose-response curve can be shifted to the right by atropine. Although acetylcholine has been shown to be effective in increasing renal blood flow in allograft rejection in dogs (Hollenberg et al. 1968b), information regarding its effects in established acute renal failure is lacking. An attempt therefore has been made to determine the effect of acetylcholine on renal hemodynamics and renal func-

tion in established acute renal failure induced by noradrenaline in the dog.

a. Methods:

Five mongrel dogs of either sex were used. The general experimental procedure was as outlined in Section III, B, with the following modifications: Prior to induction of acute renal failure by noradrenaline, acetylcholine hydrochloride was infused into the right kidney at rates in multiple increments of 48.5 ug/min calculated as weight of the base until a maximum response in total renal blood flow was obtained. At 2 and 6 hours post-noradrenaline infusion, graded rates of acetylcholine infusion were repeated. Urine samples were collected for clearance studies during infusion of acetylcholine.

The effect of intrarenal acetylcholine infusion on renal hemodynamics was also studied in one of the chronic experimental animals (Section III, B Dog C-5) by selective renal angiography and Xenon^{133} washout 8 days post-noradrenaline infusion.

b. Results:

Infusions of acetylcholine into the right renal artery resulted in marked increases in total renal blood flow in a dose-related fashion. Maximum increases in renal blood flow, expressed as a percentage of renal blood flow immediately prior to acetylcholine infusion, are shown in Table XXI. No significant differences could be observed in the control period, and at 2 hours or 6 hours post-noradrenaline infusion.

TABLE XXI

Effect of Acetylcholine on Renal Blood Flow
in Noradrenaline-induced Acute Renal Failure
in the Dog

	Control Period	6 Hours Post-NA Infusion	P value
Max. RBF (% Pre-Ach RBf)	195.5 (10.9)	188.1 (16.1)	NS
Mean B. P. (mm Hg)	135.0 (7.4)	133.8 (6.2)	NS

NS = Not significant

Non-paired t test used for statistical analysis

Both urine flow rates and sodium excretion increased during acetylcholine infusion. However, no significant changes in endogenous creatinine clearance were observed.

The renal vascular response to I. A. acetylcholine in a typical experiment is shown in Figure 13. All experimental animals demonstrated qualitatively similar vascular responses. The dose-response curve of % change in RBF versus log-dose of intra-arterial acetylcholine is shifted to the right with the development of acute renal failure although qualitatively responses were identical to that seen in the normal kidney. Concomittant I. A. infusion of atropine at a rate of 4.85 ug/min greatly attenuated or completely abolished the vasodilating effects of acetylcholine.

The results of the renal vascular response to a 5 minute-infusion of acetylcholine at a rate of 197 ug/min in an animal 8 days post-noradrenaline infusion are shown in Table XXII. There was no significant difference in the fractional increase in mean renal blood flow and pattern of distribution between the normal left kidney and the diseased right kidney. Figures 14 and 15 demonstrate the abnormal angiographic appearance of the right kidney and the normal appearance of the left kidney before and after intra-arterial acetylcholine infusion. An increase in calibre of all renal vessels visualized occurred with acetylcholine infusion.

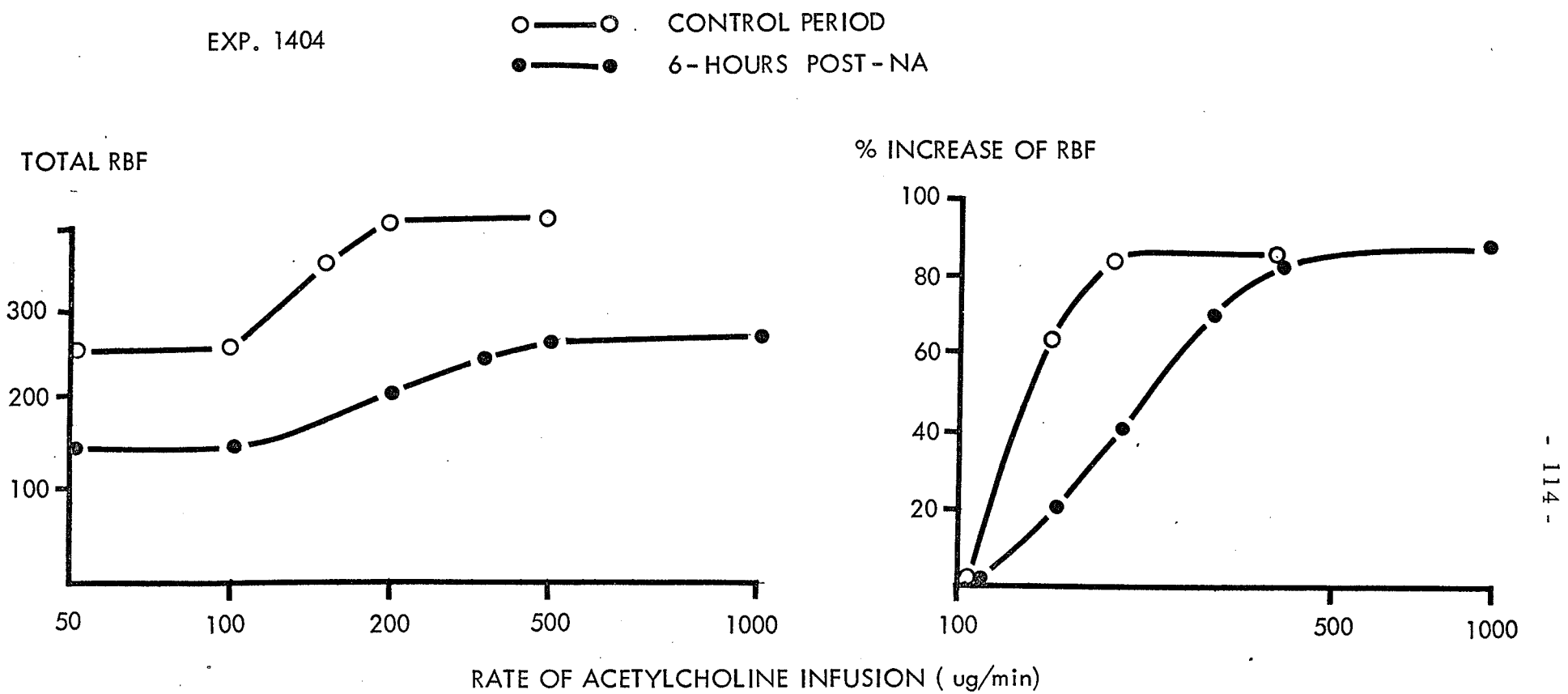


Figure 13: Effect of acetylcholine on renal blood flow in noradrenaline-induced acute renal failure in the dog.

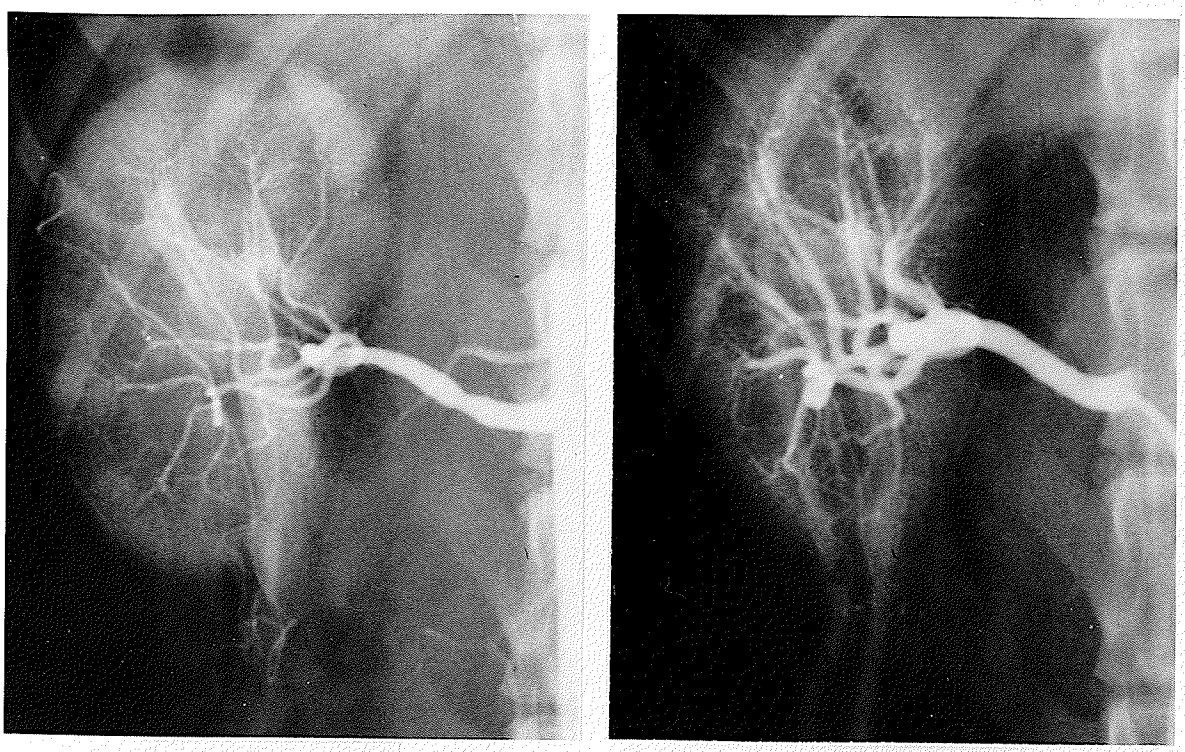


Figure 14: Angiographic appearance of right kidney before and after acetylcholine in noradrenaline-induced acute renal failure (Exp. C-5).
Diminished cortical perfusion of the right kidney 8 days post-noradrenaline infusion was evident both before and after acetylcholine despite the marked increase in calibre of the more proximal branches of the right renal artery.

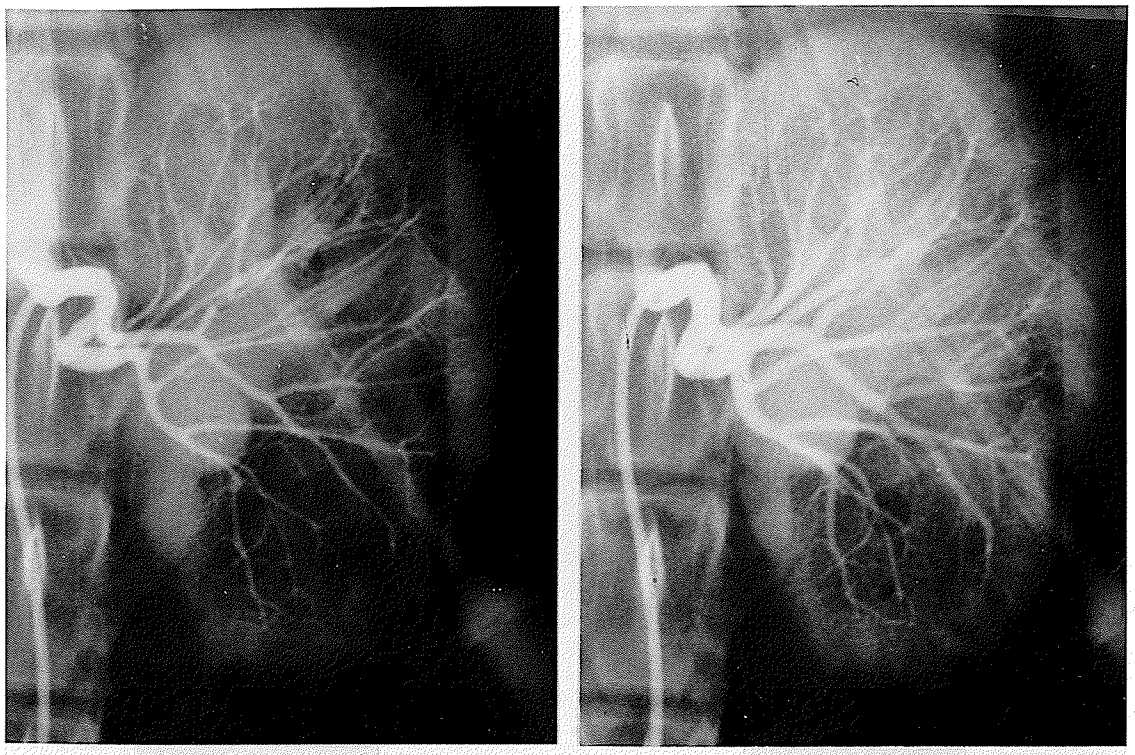


Figure 15: Angiographic appearance of normally functioning left kidney before and after acetylcholine (Exp. C-5). Normal renal vasculature before acetylcholine; marked increases in calibre of more proximal vessels following acetylcholine infusion.

TABLE XXII

Effect of Acetylcholine on IRBD in Noradrenaline-induced Acute Renal Failure (experiment C-5)

		RBF ml/100 gm/min			% RBF		
		C_I	C_{II}	Mean	C_I	C_{II}	$C_{III + IV}$
Left	Before ACh	392.0	104.0	218.5	64.0	25.0	11.0
	After ACh	522.5	188.2	375.2	70.0	26.0	4.0
Right	Before ACh	535.5	95.9	139.7	36.5	49.9	14.5
	After ACh	850.1	153.3	240.0	32.0	45.2	22.8

c. Comments:

The renal vasodilating properties of acetylcholine have been well documented. Angiographic appearance of the renal vasculature following intra-arterial acetylcholine, as shown in Figures 14 and 15, is consistent with the findings of Freed et al. (1969). It is apparent that in established noradrenaline-induced acute renal failure in the dog, the renal vasculature responds in a fashion qualitatively similar to that seen in the normal kidney. Atropine was able to block the response to acetylcholine both before and after noradrenaline infusion. This confirms the observations previously reported by other investigators in normal dogs (Vander, 1965; McGiff and Burns, 1967 and DiSalvo and Fell, 1971). No increase in endogenous creatinine clearances were observed during vasodilatation. One of the explanations offered is that a proportionate decrease in post-glomerular resistance relative to pre-glomerular resistance occurs with acetylcholine infusion (Carriere et al. 1971). Although no tachyphylaxis to acetylcholine was observed during the period of infusion, the vasodilating effects were relatively short-lived and returned to pre-infusion values with the cessation of infusion.

In summary, the present studies showed that in established acute renal failure, intra-arterial acetylcholine is still capable of increasing renal blood flow. The inability to achieve a complete hemodynamic recovery is consistent with the theory that abnormal mediators

are present and acting as physiological antagonists, or that only a portion of the renal vascular bed is available for such manipulation or that there is an alteration in the base-line reactivity of the vascular smooth muscle. This situation is in contrast to that seen in early renal allograft rejection in the dog where I. A. acetylcholine could achieve the same maximum renal blood flow before rejection set in (Hollenberg et al. 1968b).

(ii) Renal Vascular Responses to Phenoxybenzamine

The pattern of intrarenal vascular response to catecholamine infusion (Carriere, 1969; and Epstein et al. 1970b) or to hemorrhagic hypotension (Carriere et al. 1966; Bell and Harper, 1968) has been found to be qualitatively similar to that seen in established renal failure (Hollenberg et al. 1968a, 1970). Schnermann et al. (1966) suggested that renal ischemia resulted in the continued local activation of the renin-angiotensin system. The initiating event which causes the renal ischemia, whether it be shock and sympathetic nervous system overactivity (Hollenberg, 1965), the direct vascular effects of toxins (Oliver, 1953) or the vascular effects of an immunologic process (Hollenberg et al. 1968b), could be responsible for the subsequent sustained cortical ischemia. This is consistent with our finding that prior renal denervation protected the rat from acute renal failure. However, it is uncertain as to when the sympathetic influences fail to be rate-limiting. The

time of post-event phenoxybenzamine blockade in determining eventual renal responses in hemorrhagic shock is critical (Hollenberg, 1965). We have demonstrated that intra-peritoneal administration of phenoxybenzamine 2.5 hours post-glycerol injection partially protects the rat from acute renal failure (Section III, A). However, the duration of renal insult following glycerol injection as reflected by the occurence of hemoglobinuria may last up to 12 hours. Therefore, attempts were made to determine the qualitative contribution of adrenergic influences in established acute renal failure in the dog.

a. Methods:

Five mongrel dogs of either sex, weighing 15-30 kg were used. The experimental procedure was as outlined in Section III, B with the following modifications: Phenoxybenzamine at doses of 0.2 mg/kg B.Wt. (calculated as the base) was infused directly into the right renal artery over 10 min by a constant infusion pump 6 hours post-noradrenaline infusion. At this time, mean total renal blood flow was usually 40% of control. Adequacy of adrenergic blockade was assessed by the renal vasoconstrictor response to brief intra-arterial noradrenaline before POB and a complete lack of vasoconstriction in the presence of systemic rise in arterial blood pressure after POB. Renal function and total renal blood flow of the right kidney were measured for 30 minutes post-POB blockade. These were compared to the

corresponding parameters in 12 non-POB treated controls (Section III, B, 2 (ii)). The effect of intrarenal phenoxybenzamine infusion on renal hemodynamics was also studied in one of the chronic experimental animals (Section III, B Dog C-6) by selective renal angiography and Xenon^{133} washout 24 hours post-noradrenaline infusion.

b. Results:

Results are shown in Table XXIII. Intra-arterial administration of phenoxybenzamine was not associated with statistically significant changes in renal hemodynamics and renal function 6 hours post-infusion of noradrenaline when compared to non-POB treated controls. Mean B.P. remained unchanged during the experimental period.

In experiment C-6, 24 hours post-noradrenaline infusion to the right kidney, phenoxybenzamine at a dose of 0.2 mg/kg was infused over 10 minutes into left and right kidneys. Selective renal arteriography and Xenon^{133} washout studies were performed before and after the administration of phenoxybenzamine. Figures 16 and 17 demonstrate the renal angiographic appearances of experiment C-6 before and after phenoxybenzamine. The right renal vasculature showed considerable diminished cortical perfusion. Following the administration of phenoxybenzamine, increase in vessel calibre down to the interlobar level was observed. No increase in cortical perfusion could be appreciated. The normally functioning left kidney showed normal angiographic appearances

TABLE XXIII

Effect of Phenoxybenzamine on Renal Blood Flow in Noradrenaline-induced Acute Renal Failure in the Dog

	Non-treated Controls N=9	Phenoxybenzamine-treated N=4	p Values
RBF (% Control)	0.01 ± 0.01	0.00 ± 0.01	NS
B.P. (mm Hg)	1.5 ± 0.82	0.8 ± 0.32	NS
$\dot{V}$ (% Control)	21.57 ± 4.61	23.22 ± 6.6	NS
C_{Cr} (% Control)	6.4 ± 1.1	4.2 ± 1.4	NS

Results shown are Mean (S.E.) of the absolute values of the differences between the period 4-6 hours and 6.0-6.5 hours post-NA infusion in non-POB pretreated controls and in POB pretreated animals.
Non-paired t test was used for statistical analysis.

both before and after phenoxybenzamine. Similar to the right kidney, dilatation can be seen in the more proximal branches of the left renal artery following phenoxybenzamine. Both nephrogram phases remained unchanged.

Table XXIV shows mean RBF and IRBD before and after phenoxybenzamine as assessed by $Xenon^{133}$ washout. No changes were present before and after phenoxybenzamine in both kidneys. Despite the increase in calibre of the more proximal vessels, mean RBF remained low post-phenoxybenzamine administration. The fast-flowing component C_I remained unidentifiable.

c. Comments:

Unlike the glycerol-rat model, the duration of renal insult in the noradrenaline-dog model is more sharply demarcated. The direct pharmacologic effect of noradrenaline is probably over within a few minutes after noradrenaline infusion has been stopped as evidenced by the rapid return of the elevated systemic blood pressure, seen during infusion, to control values. This feature makes the noradrenaline-dog model particularly suitable for studying adrenergic influences during the persistent renal vasoconstriction seen in established acute renal failure.

Intra-arterial phenoxybenzamine given at a dose of 0.2 mg/kg B.Wt. was sufficient to block the renal vascular effects of exogenous noradrenaline, but not high enough to produce significant changes in

systemic hemodynamics. This dose has been shown to be adequate in completely preventing the adverse effects of prolonged I. A. noradrenaline infusion (Section III, B. Exp. 4). The lack of effect of intra-arterial phenoxybenzamine at this dosage on renal hemodynamics in the post-noradrenaline infusion period is consistent with the theory that adrenergic influences are not significantly involved in the persistent renal cortical ischemia seen in established acute renal failure.

In Dog C-6, 24 hours post-noradrenaline infusion, angiography showed an increase in calibre of the more proximal branches of the right artery with phenoxybenzamine blockade. However, no change in calibre of higher order vessels or the nephrogram was observed. These findings were supported by the $Xenon^{133}$ washout data where no obvious changes in mean renal blood flow or redistribution of intrarenal blood flow were documented. This is consistent with the hypothesis that the increase in renal vascular resistance is at the level of the preglomerular vessels. The increase in large vessel calibre with phenoxybenzamine blockade suggests that considerable adrenergic vascular tone existed in vessels of this size at the time of experimentation. This was likely a function of the experimental conditions at the time since the contralateral normally functioning left kidney demonstrated qualitatively the same response.

On the basis of the above experiments, it can be stated that

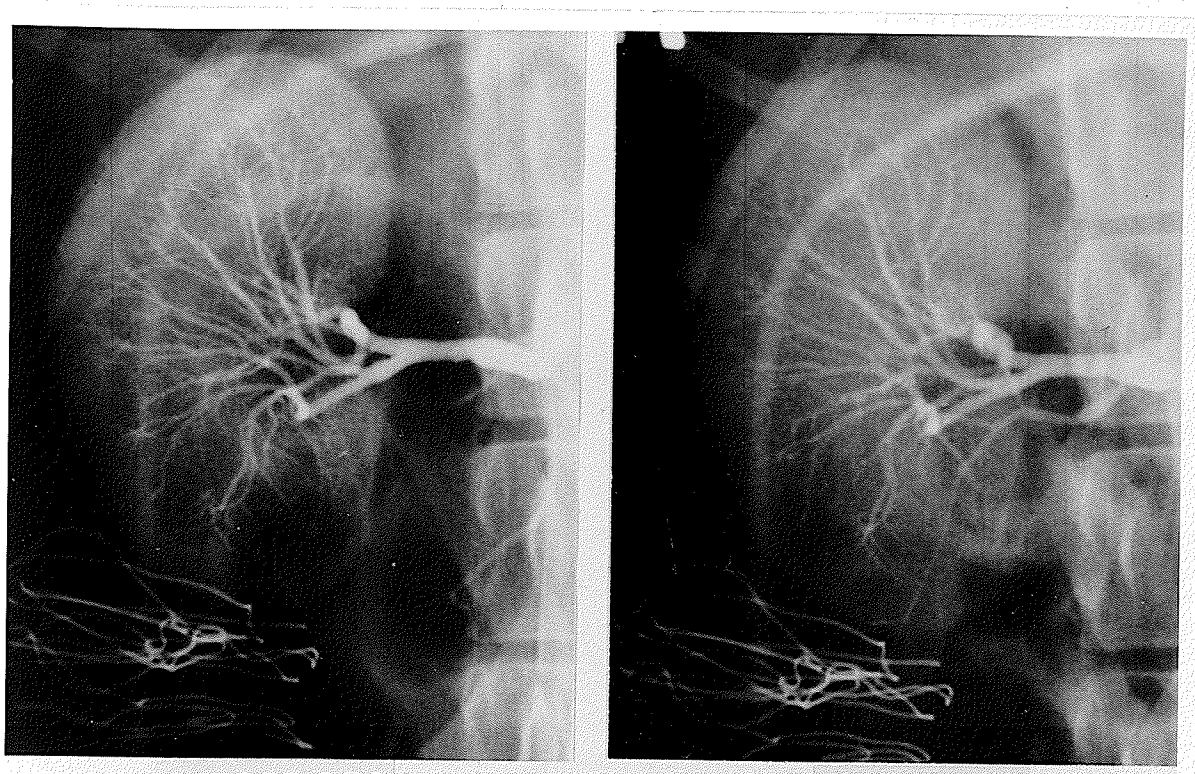


Figure 16: Angiographic appearance of right kidney before and after phenoxybenzamine (Exp. C- 6). Diminished cortical perfusion of the right kidney 24 hours post- noradrenaline infusion before and after phenoxybenzamine despite some degree of vasodilatation in the more proximal branches of the right renal artery.

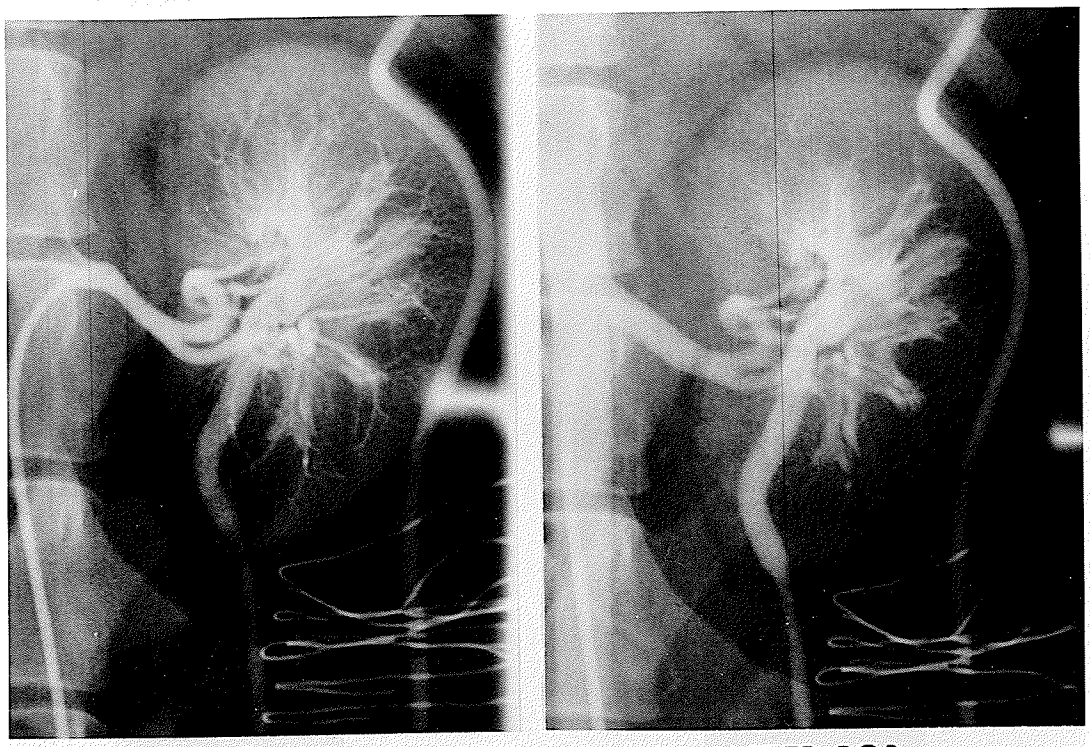


Figure 17: Angiographic appearance of normally functioning left kidney before and after phenoxybenzamine (Exp. C-6).
Normal renal vasculature can be observed before the administration of phenoxybenzamine. The more proximal branches of the left renal artery showed increase in calibre following phenoxybenzamine administration.

TABLE XXIV

Effect of Phenoxybenzamine on IRBD in Noradrenaline-induced Acute Renal Failure in the Dog (experiment C-6)

	RBF ml/100 gm/min			% RBF		
	C_I	C_{II}	Mean	C_I	C_{II}	$C_{III + IV}$
Left Kidney Before POB	714.0	100.4	459.0	75.0	18.0	7.0
Left Kidney After POB	845.5	89.2	535.5	76.0	8.5	15.5
Right Kidney Before POB		459.0	128.5		38.4	61.6
Right Kidney After POB		440.0	110.8		38.2	61.8

both the normal kidney and the hypoperfused kidney with depressed renal function 24 hours post-noradrenaline infusion demonstrated similar adrenergic vascular responses, viz., vasoconstriction to noradrenaline and little change with phenoxybenzamine blockade.

(iii) Effect of Acute Volume Expansion

Prolonged infusions of noradrenaline cause plasma volume depletion (Sutter 1962). Despite generous intravenous fluid replacement (Section III, B, (i)) central venous hematocrit rose considerably during the infusion period with gradual return to control values. It is appreciated that at least part of the diminished renal blood flow observed even at 6 hours post-noradrenaline infusion might be due to the effect of plasma volume depletion. In this experimental situation, an attempt was made to assess the effect of acute volume expansion on renal blood flow.

a. Methods:

Five mongrel dogs of either sex were used. The experimental procedure was as outlined in Section III, B with the following modifications: Acute volume expansion was accomplished by intravenous administration of 0.9% saline at a rate of 25 ml/kg B.Wt. given over 30 minutes at 6 hours post-noradrenaline infusion. Renal hemodynamics and function during the period of volume expansion was compared to 12 non-treated controls.

b. Results:

Results are shown in Table XXV. Acute volume expansion was associated with significant but variable increases in total renal blood flow and urine flow rates in all animals. Systemic blood pressure remained unchanged. Minor but non-significant increases in creatinine clearances were observed.

c. Comments:

As expected, in the hypovolemic animal, acute volume expansion was associated with definite increases in total renal blood flow without significant changes in blood pressure. Again, the renal vascular response was appropriate for the stimulus.

The minor improvement in GFR observed together with the fact that increase in urine flow rates occurred in 4 out of 5 cases suggests that the renal vasoconstriction was not homogenous. A small percentage of the nephrons somehow escape the damaging effect of the insult and pre-glomerular resistance relative to post-glomerular resistance was low enough that filtration pressure was maintained. This implies a patchy distribution of the "functional" lesion. Such a phenomenon would support the speculation that a differential response of the vascular smooth muscle to the insult exists, probably in a gaussian fashion.

TABLE XXV

Effect of Volume Expansion on Renal Blood Flow in Noradrenaline-induced Acute Renal Failure in the Dog

	Non-treated Controls N=9	Acute Volume Expansion N=4	p Value
RBF (% Control)	0.01 ± 0.01	73.2 ± 5.3	<0.001
B.P. (mm Hg)	1.5 ± 0.82	0.0 ± 1.4	NS
$\dot{V}$ (% Control)	21.57 ± 0.61	272.5 ± 107.5	<0.001
C_{Cr} (% Control)	6.4 ± 1.1	5.4 ± 0.4	NS

Results are expressed as Mean (S.E.) of the differences between the period 4-6 hours and 6.0-6.5 hours post-NA infusion in non-treated controls and in animals receiving acute volume expansion.
Non-paired t test was used for statistical analysis.

(iv) Discussion

The foregoing studies showed that in established acute renal failure, the renal vascular bed, although functioning at an abnormally low base-line, is capable of responding to pharmacologic and physiologic stimuli in an appropriate fashion, qualitatively similar to the behavior of the normal kidney. This implies the qualitative intactness of the effector mechanism, and in particular, the cholinergic and adrenergic mechanisms. The inability to achieve complete return of renal blood flow to control by acetylcholine while noradrenaline was able to decrease renal blood flow (just before POB blockade 6 hours post-noradrenaline infusion) may be explained by either a decrease in available receptors (drug unable to reach receptor site due to already existing marked vasoconstriction) or a decrease in "intrinsic activity" of the vascular smooth muscle to dilate but not to contract, or a combination of the above. The possibility of physiological antagonism by other mediators cannot be ruled out.

2. Early Acute Oliguric Renal Failure in Man.

(i) Renal Vascular Responses to Intra-arterial Phenoxybenzamine Blockade

Enhanced adrenergic activity in clinical shock of mixed etiology has been implicated as being an important factor for the production of oliguria and renal failure (Thomson, 1965). However, information is lacking regarding the contribution of the adrenergic nervous system in the persistent renal vasoconstriction once acute renal failure is well established. We attempted to determine the effect of intrarenal phenoxybenzamine blockade on renal hemodynamics in established acute oliguric renal failure in the presence of stable systemic hemodynamics in man.

a. Choice of Patients

Patients with unequivocal clinical evidence of acute oliguric renal failure were chosen for this study. The criteria of established acute renal failure includes: oliguria, with urine flow rates of less than 30 ml/hr for a minimum of ten hours; urinary sodium concentration greater than 50 mEq/l in presence of oliguria; azotemia and lack of renal response to volume expansion.

Four cases of acute oliguric renal failure (ages 43-78) were studied. In addition, one case of acute oliguric post renal allograft transplant due to graft rejection and one case of normal renal function

TABLE XXVI

Clinical Background of Patients Studied

Patient	Age	Sex	Clinical Diagnosis	Duration of Oliguria before study	BUN (mg%) at time of POB Blockade	U_{Na} mEq/l before study
G. L. WGH 203635	78	male	perforated sigmoid colon, sepsis, pulmonary embolism, acute renal failure.	24 hrs	68	56
E. G. WGH 354046	74	male	first and second degree body burns, pneumonia, acute renal failure	3 days	100	63
J. B. WGH 308048	48	male	first and second degree body burns, shock, acute renal failure.	12 hrs	40	55
I. C. WGH 282623	43	female	septic shock, pseudomonas pneumonia, peritonitis, infarction of terminal ileum, acute renal failure.	30 hrs	94	88
D. R. WGH 184326	26	male	renal allograft rejection 26 days post-transplant.	36 hrs	68 (on dialysis)	80
D. K. WGH 211678	54	female	labile hypertension. Investigation for pheo-chromocytoma.	-	8	-

were studied for comparison. Clinical background for the various cases are summarized in Table XXVI.

b. Methods

In all patients studied, indwelling Foley catheters had been inserted. When the clinical diagnosis of established acute renal failure was made, and in the presence of stable systemic hemodynamics, selective renal arteriography was performed using the Seldinger technique under fluoroscopy. The renal vasculature was assessed by arteriography qualitatively and renal hemodynamics by the $Xenon^{133}$ washout method. These were repeated following the intra-arterial administration of Dibenzyline^{R} (Phenoxybenzamine hydrochloride-SKF) at a dose of 0.2 mg/kg B. Wt. given over a 10-15 minute period by a Harvard constant infusion-withdrawal pump. The adequacy of phenoxybenzamine blockade was not tested by assessing post-infusion vascular responses to adrenaline due to the clinical circumstances. This dose of phenoxybenzamine, however, when given as a continuous slow infusion, has been shown to be effective in blocking the marked vasoconstricting effects of 16 ug bolus of intra-arterial adrenaline in patient D.K. whose renal function was normal.

Angiographic appearances were assessed by two experienced radiologists one of whom was not involved in the study. Analysis of the $Xenon^{133}$ washout data has been discussed in Section III, B. Data thus obtained are compared to values obtained from human subjects

with normal renal function (n = 16) at the Winnipeg General Hospital.

Systemic arterial blood pressure and pulse rates were monitored throughout the entire experimental procedure. No significant changes in these parameters were noted before, during and immediately after the administration of phenoxybenzamine. Renal function was closely followed following blockade. One patient (Patient I. C.) received systemic phenoxybenzamine as an adjunct in shock therapy.

c. Results

Renal hemodynamic data before and after the administration of intra-arterial phenoxybenzamine are shown in Tables XXVII and XXVIII and in Figure 18 Cortical ischemia was present in all cases of established acute renal failure and in renal allograft rejection. The fast flowing component C_I seen in normal kidneys is no longer separable from the normally slower component C_{II}, by graphical analysis. Selective renal arteriograms showed poor cortical perfusion and extremely faint or absent nephrograms. No significant differences in intrarenal blood distribution or mean renal blood flow could be demonstrated by the $Xenon^{133}$ washout method post-phenoxybenzamine administration. All $Xenon^{133}$ washout tests carried out were technically satisfactory. A typical washout curve is shown in Figure 19 (Patient E. G.). Following intra-arterial phenoxybenzamine, the more proximal branches of the renal arteries showed marked dilatation in all cases of acute renal failure angiographically.

TABLE XXVII

Effect of Phenoxybenzamine on IRBD in Acute Renal Failure in Man

		RBF ml/100 gm/min			% RBF		
		C_I	C_{II}	Mean	C_I	C_{II}	$C_{III + IV}$
G. L.	Before POB	668.3	83.5	92.8	9.5	48.9	51.0
	After POB	477.4	77.7	104.4	16.6	58.2	25.0
E. G.	Before POB	98.4		94.2	72.7		27.3
	After POB	99.1		96.9	78.4		21.6
J. B.	Before POB	88.8		60.4	77.2		22.8
	After POB	77.5		53.0	71.1		28.9
Normal Values (WGH) N=9		417.1 (11.3)	115.9 (11.7)	318.2 (13.2)	72.4 (2.1)	18.1 (2.5)	9.5 (1.3)

TABLE XXVIII

Effect of Phenoxybenzamine on IRBD in Renal Allograft Rejection and in Normal Renal Function in Man

		RBF ml/100 gm/min			% RBF		
		C_I	C_{II}	Mean	C_I	C_{II}	C_{III+IV}
D.R. (Rejection)	Before POB	126.4		99.8	77.0		23.0
	After POB	114.9		88.2	78.2		21.8
D.K. (Normal)	Before POB	398.1	59.0	213.8	65.4	16.8	17.8
	After POB	354.0	65.7	205.5	64.0	21.0	15.0
Normal Values (WGH) N=9		417.1 (11.3)	115.9 (11.7)	318.2 (13.2)	72.4 (2.1)	18.1 (2.5)	9.5 (1.3)

No changes in calibre of higher order vessels could be seen. Nephrogram remained poor. A typical example of the angiographic appearance before and after POB administration is shown in Figure 17 (Patient E. G.). No improvement of renal function occurred following POB treatment.

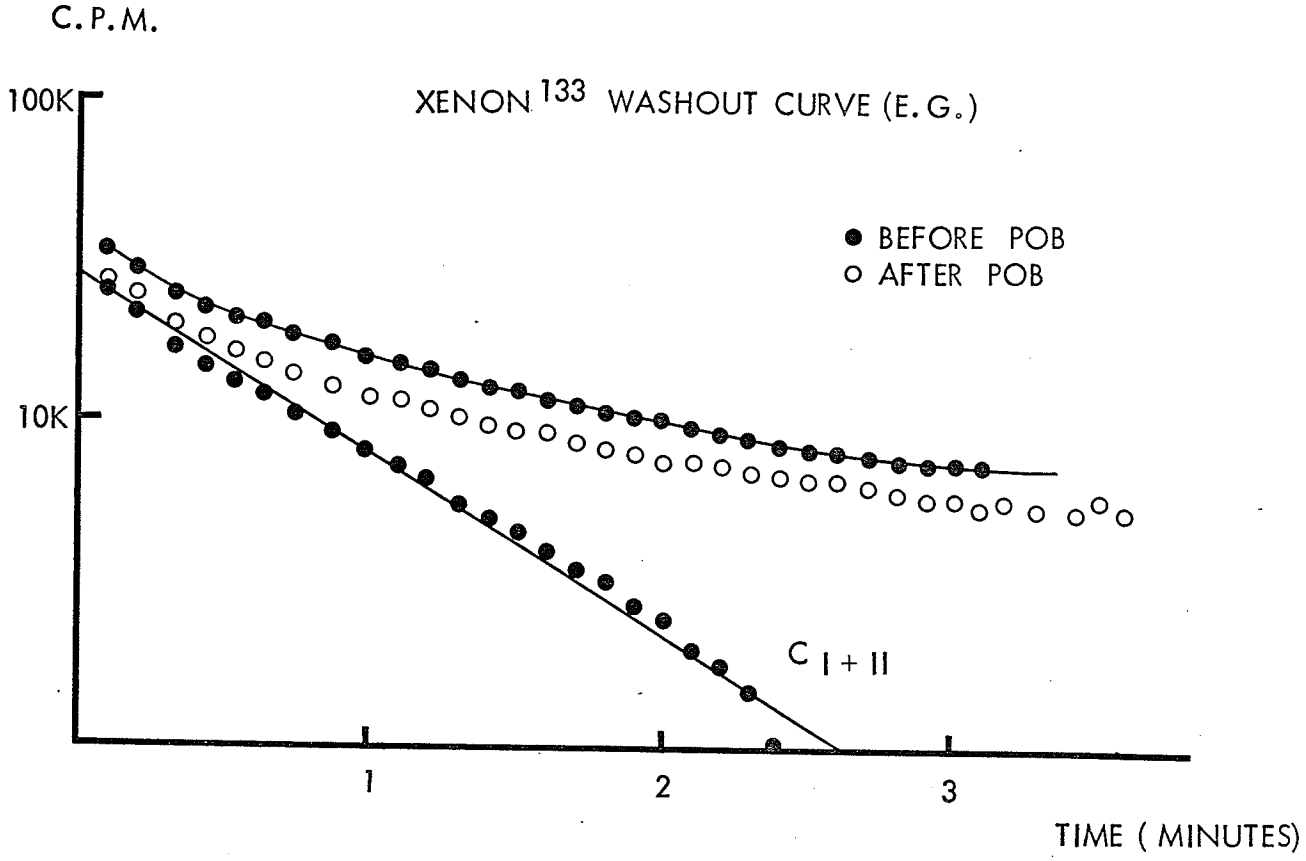


Figure 18: Effect of phenoxybenzamine on IRBD in acute renal failure in man (Subject E. G.).

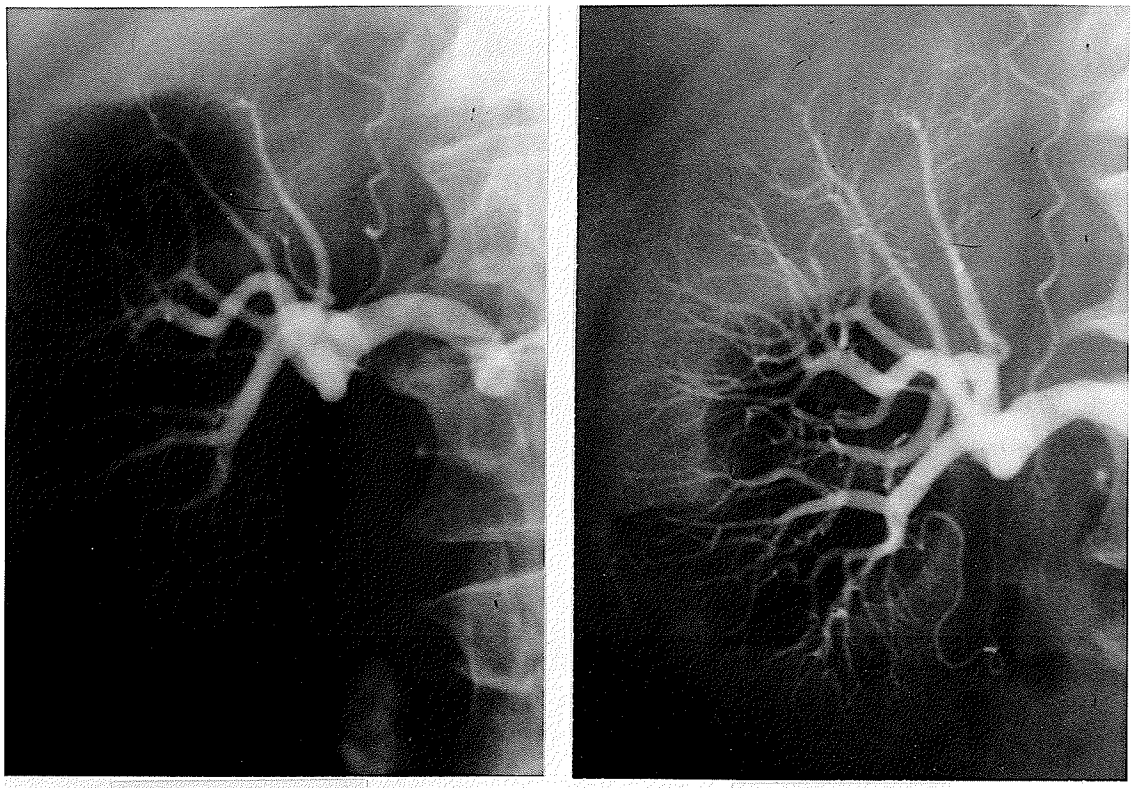


Figure 19: Angiographic appearance of right kidney before and after phenoxybenzamine in acute renal failure in man (Subject E. G.). Diminished cortical perfusion, typically seen in acute renal failure, is evident both before and after phenoxybenzamine administration despite marked dilatation of the proximal branches of the right renal artery.

d. Discussion

Renal cortical ischemia was present in all of the four cases studied where acute renal failure was well established. This is supported by the angiographic findings described and by the absence of a fast flow component C_I on $Xenon^{133}$ washout. At a time when systemic hemodynamics were stable, the intra-arterial administration of phenoxybenzamine at a dose low enough not to alter systemic blood pressure resulted in dilatation of the more proximal branches of the renal artery but not in vessels beyond the arcuate level. No concomitant improvement in $Xenon^{133}$ washout curves were seen. This pattern of response, namely: dilatation of more proximal vessels and lack of response in smaller vessels is strikingly similar to that seen in the acute renal failure kidney in the dog and in the normal kidney in both the dog and in man.

The dose of phenoxybenzamine used has been demonstrated to be adequate in preventing the vascular effects of subsequent intra-arterial noradrenaline at very high doses in the dog (Section III, Exp. 2 (ii)) and in one case in the normal human. The use of bolus injections of intra-arterial noradrenaline or adrenaline for the assessment of adequacy of alpha adrenergic blockade cannot be used in oliguria for obvious ethical reasons. However, it is likely that a considerable degree of alpha blockade was achieved.

The richness of adrenergic fibers innervating the preglomerular arteries (McKenna and Angelakos, 1968) would suggest that if adrenergic influences are significantly responsible for the renal vasoconstriction, this would be the likely site of action. The lack of improvement in renal hemodynamics following the administration of phenoxybenzamine supports the theory that adrenergic mediation is not responsible for the cortical ischemia in established acute renal failure. The angiographic observation that the larger branches of the renal artery dilated following the phenoxybenzamine treatment implies the presence of some sympathetic tone in these Windkessel vessels and that the dose used did produce a pharmacologic effect.

It has been suggested by Hollenberg et al. (1968b) that the ischemia in early rejection may be a reversible phenomenon. Since adrenergic denervation is complete in the transplanted kidney (Almgard et al. 1971), the lack of dilatation even in larger vessels in oliguria due to allograft rejection post-POB is consistent with the theory that the adrenergic influences plays little part in the ischemia observed. The fact that acute tubular necrosis occurs post-renal transplantation is also consistent with our suggestion that the adrenergic nervous system is not responsible for the persistent renal vasoconstriction in established renal failure. However, it is interesting to note that again the diseased kidney and the normal kidney responded in a similar way.

SECTION IV

GENERAL DISCUSSION AND CONCLUSIONS

The present study has shown that a 2 hour infusion of noradrenaline, directly into one renal artery of a dog, can result in a lesion closely resembling that seen in human cases of acute renal failure of varied etiology. In this model, only the renal effects of noradrenaline infusion were persistent beyond the period of infusion. The presence of a normally functioning contralateral kidney permitted the sequential study of intrarenal events essentially unmodified by the systemic consequences of renal failure per se. In this model it was found necessary to have almost complete cessation of renal blood flow during the infusion of noradrenaline in order to induce renal failure. When renal blood flow was preserved, by prior alpha adrenergic blockade with phenoxybenzamine, the characteristic changes in intrarenal hemodynamics and in function were not seen. These findings support the proposal that the induction of acute renal failure can be a purely vascular event.

The importance of the sympathetic nervous system and its relationship to chronic salt loading, in the development of acute renal failure, were shown by the experiments using the glycerol-induced rat model. The findings of these studies are in general agreement with those of other authors, both in humans and in other animal models. Powers et al. (1957) demonstrated that prior renal denervation or ganglionic blockade in man decreased the renal damage due to cross-

aortic clamping in aneurysectomies. Ruiz-Guinazu (1971) reported that prior systemic alpha adrenergic blockade by phenoxybenzamine prevented the development of acute renal failure following the administration of methemoglobin directly into the renal artery in the dog. Systemic and intra-arterial administrations of phenoxybenzamine before harvest of cadaveric donor kidneys were found to be associated with a reduced incidence of immediate post-transplant acute tubular necrosis (Thomson, personal communication). The richness of adrenergic innervation (McKenna and Angelakos, 1968) and the lack of resting sympathetic tone (Smith et al. 1939) in the kidney render the renal circulation particularly vulnerable to situations of prolonged stress. Other vasoactive systems, e.g. the renin-angiotensin axis; and other "protective" pretreatment manuevers, e.g. prior volume expansion, chronic salt loading, etc., may play only a permissive role in the development of acute renal failure. These findings strongly support the theory that activation of the sympathetic nervous system was an important step in the initial development of acute renal failure. Whether the sympathetic nervous system is equally important in the initial production of acute renal failure due to nephrotoxins is unclear. A vascular phenomenon may also be responsible. The two recently described toxin-induced acute renal failure models in the dog, involving the use of mercury chloride (Solomon et al. 1971) and uranyl nitrate (Flamenbaun

et al. 1971), have been demonstrated to be associated with cortical ischemia similar to the vascular changes found in the noradrenaline-dog model and in human cases. These models may prove to be useful for the study of sympathetic involvement in nephrotoxin-induced acute renal failure.

The cause of the persistent cortical ischemia in acute renal failure is still open to speculation. We have demonstrated that alpha adrenergic blockade with phenoxybenzamine in established acute renal failure in the dog and in man was not associated with improvement of intrarenal hemodynamics and GFR. This, together with the occurence of acute renal failure in renal transplants (Almgard et al. 1971) strongly suggests that the increased renal vascular resistance in both innervated and transplanted kidneys in established acute renal failure could not be due to a persistence of the elevated sympathetic tone. Moreover, it is difficult to envision a unilateral increase in sympathetic activity being responsible for the maintenance of unilateral renal failure in the noradrenaline-dog model.

The theory first advanced by Goormaghtigh (1945, 1947) and subsequently modified by others (Sevitt, 1959; Finckh, 1962; Schnermann et al. 1966; Henry et al. 1968; Brown et al. 1970), proposed that the renin-angiotensin system was responsible for the persistence of renal ischemia. However, hypoxemia per se has not been shown to be a

stimulus for renin release (Skinner et al. 1963) and the decrease in renal blood flow observed in the dog after breathing an hypoxic gaseous mixture has been found to be abolished by renal denervation (Korner, 1963). These findings do not support the thesis that the persistant intrarenal release of renin was secondary to hypoxia in established acute renal failure. The likelihood of the renin-angiotensin system being responsible for the maintenance of renal vasoconstriction has been critically discussed in Section III.

Other oliguric states, e.g. renal allograft rejection and the hepato-renal syndrome, are also characterized by reduced renal blood flow predominantly involving the outer cortex. A number of important features serve to distinguish these oliguric disorders from classic acute renal failure. Although it has been suggested that a potentially reversible vascular phase exists early in allograft rejection (Hollenberg et al. 1968b), precisely how this is related to ongoing immunologic events is unknown. Although it has been demonstrated that prior phenoxybenzamine blockade could prevent the Schwartzman reaction (Muller-Bergaus and McKay, 1967; Muller-Bergaus et al. 1967), inhibit the activation of Hageman factor (McKay et al. 1970), it would be erroneous to imply that the alpha adrenergic mediator was responsible and that phenoxybenzamine can influence graft rejection.

In the hepato-renal syndrome, an unstable pattern of cortical

ischemia has been demonstrated by the Xenon^{133} washout technique (Epstein et al. 1970). This "instability" of the Xenon^{133} washout curve in the most oliguric cases, has been attributed to the presence of some abnormal mediator being responsible for the observed cortical ischemia. The inability of vasodilating agents, including phentolamine and acetylcholine to improve blood flow suggest respectively a lack of sympathetic component in the observed vasoconstriction and the possibility of marked physiologic antagonism. Both of the above oliguric states have one feature in common. This is the continued presence of the initiating insult. Removal of the triggering events, e.g. by treatment of rejection, or the transplantation of the kidneys from patients with hepatorenal syndrome to recipients with normally functioning livers would result in prompt improvement in intrarenal hemodynamics and function.

In contrast, acute renal failure is characterized by the persistence of depressed function and disturbances in intrarenal hemodynamics when the initial insulting event has long ceased to be present. Based on the rapidly increasing body of knowledge regarding vasoactive substances and the intrarenal circulation (reviews by: Hatch and Johnson, 1969; Barger, 1971; Grunfield et al. 1971; Thurau and Levine, 1971), emphasis of current speculations on the nature of the persistent renal vasoconstriction has been directed towards the continued local release of some vasoconstrictor. Sweet et al. (1971) reported the presence of

a substance enhancing vascular responsiveness to catecholamines following acute renal ischemia. Hinshaw (1971) reported altered patterns of autoregulation during shock and renal ischemia. However, all of these observations were made in acute experimental conditions. Several other findings tend to cast some doubt on this explanation. The oliguric phase in acute renal failure may last weeks. The lack of development of tachyphylaxis to the postulated mediators, the short transitional period between the oliguric and diuretic phases, and the stability of the pattern of intrarenal blood flow distribution reflected by reproducibility of $Xenon^{133}$ washout curves over a period of several days, would argue against the presence of mediators being responsible for the maintenance of cortical ischemia. In addition, in the absence of the initial insulting stimulus, a positive feedback mechanism would have to be postulated. This theory does not explain the high potential for eventual functional recovery.

In contrast to renal vascular responses seen in canine allograft rejection (Hollenberg et al. 1968b) or in the hepato-renal syndrome (Epstein et al. 1971), we have demonstrated that the vascular responses to acetylcholine and phenoxybenzamine are qualitatively similar in both the diseased kidney and the normal kidney. The identical maximum fractional increases in renal blood flow in both kidneys, and in the absence of gross structural lesions involving the glomeruli and the vasculature, are consistent with the theory that a persistent change in vascular re-

activity is present in established acute renal failure. The maintenance of a high pre-glomerular resistance may be due to a long lasting but reversible alteration of resting tone of the vasculature involved. This is reflected by a decrease in "intrinsic activity" of the vasculature to dilate in response to the administration of acetylcholine. This hypothesis would imply a post-receptor change in acute renal failure induced by the insult. One can only speculate on the nature of this change. It might involve subtle cell-membrane changes or a change in the contractile mechanism, the recovery of which would be associated with the improvement of intrarenal hemodynamics.

In summary:

1. A reproducible model for the study of renal hemodynamics in acute renal failure has been developed in the dog by prolonged infusion of noradrenaline directly into the renal artery.

2. The development of acute renal failure could be a purely vascular event.

3. The sympathetic nervous system probably plays an important role in the initiation of acute renal failure.

4. Activation of the renin-angiotensin system cannot be singularly responsible for the development of acute renal failure. The protective effects of salt loading could not be equated to the effects of renin depletion.

5. Enhanced sympathetic overactivity cannot be responsible for the persistence of renal vasoconstriction in established acute renal failure.

6. The renal vascular responses to acetylcholine and to phenoxybenzamine in acute renal failure are qualitatively similar in the diseased and in the normal kidney.

SECTION V

REFERENCES

Ayer, G., Grandchamp, A., Wyler, T. and Traniger, D.: "Intrarenal hemogynamics in glycerol-induced myohemoglobinuric acute renal failure in the rat". Circulation Res. 29: 128, 1971.

Baker, C.H.: "Relationships between renal vascular volume and blood flow resistance". Am. J. Physiol. 219: 1337, 1970.

Barenberg, R.L., Solomon, S., Papper, S. and Anderson, R.: "Clearance and micropuncture study of renal function in mercuric chloride treated rats". J. Lab. Clin. Med. 72: 473, 1968.

Barger, A.C., Muldowney, F.P. and Liebowitz, M.R.: "Role of the kidney in the pathogenesis of congestive heart failure". Circulation, 20: 273, 1959.

Barger, A.C.: "Renal hemodynamic factors in congestive heart failure". Ann. N.Y. Acad. Sci. 139: 276, 1966.

Barger, A.C.: "The renal circulation". New Engl. J. Med. 284: 482, 1971.

Barraclough, M.A. and Mills, I.H.: "Effect of bradykinin on renal function". Clin. Sci. 28: 69, 1965.

Bell, G. and Harper, A.M.: "Measurement of local blood flow in the renal cortex from the clearance of Krypton 85". J. Surg. Res. 5: 382, 1965.

Bell, G. and Harper, A.M.: "The effect of haemorrhagic shock on the blood flow through the renal cortex of the dog". In: Blood Flow Through Organs and Tissues. Ed. Bain, W.H. and Harper, A.M., Livingstone 1968, page 441.

Bell, G. and Lister, G.D.: "The effect of noradrenaline and phenoxybenzamine on the renal response to hemorrhage". Gynecol. and Obstet. 130: 813, 1970.

Beleslin, D., Bisset, G.W., Haldar, J. and Polak, R.L.: "The release of vasopressin without oxytocin in response to haemorrhage". Proc. Roy. Soc. London, Ser. B. 166: 443, 1967.

Biber, T. U. L., Mylle, M., Baines, A. D., Gottschalk, C. W., Oliver, J. R. and MacDowell, M. C.: "A study by micropuncture and microdissection of acute renal damage in rats". Am. J. Med. 44: 664, 1968.

Blackburn, C. R. B., Hensley, , Grant D. K. and Wright, F. B.: "Studies on intravasculer hemolysis in man: Pathogenesis of initial stages of ARF". J. Clin. Invest. 33: 825, 1954.

Block, M. A., Wakim, K. G. and Mann, F. C.: "Certain features of the vascular beds of the cortico-medullary and medullary regions of the kidney". Arch. of Pathol. 53: 437, 1952a.

Block, M. A., Wakim, K. G. and Mann, F. C.: "Circulation through kidney during stimulation of the renal nerves". Am. J. Physiol. 169: 659, 1952b.

Bock, K. D. and Gross, F.: "Renin and angiotensin tachyphylaxis". Circulation Res. 9:1044, 1961.

Bock, K. D., Lever, A. F., Brown, J. J. and Robertson, J. I. S.: "Effects of renin and angiotensin on excretion and distribution of water and salts". In: Renal Hypertension. Ed. Page, I. H. and McCubbin, J. W. Yearbook Pub. 1968, Chap. 7, page 184.

Brandfonbrener, M. and Geller, H. M.: "Effect of dibenamine on renal blood flow in hemorrhagic shock". Am. J. Physiol. 171: 482, 1952.

Braun, W. and Merrill, J. P.: "Urine fibrinogen fragments in human renal allografts" - A possible mechanism of renal injury. New Engl. J. Med. 278: 1366, 1968.

Brest, A. N. and Moyer, J. H.: "Renal Failure". Lippincott 1967.

Brooks, F. P. and Pickford, M.: "The effect of posterior pituitary hormones on the excretion of electrolytes in dogs". J. Physiol. 142: 468, 1958.

Brown, J. J., Davis, D. L., Lever, A. F., Parker, R. A. and Robertson, J. I. S.: "Assay of renin in single glomeruli". Lancet 2: 668, 1963.

Brown, J. J., Davies, O. L., Lever, A. F. and Robertson, J. I. S.: "Influence of sodium deprivation and loading on the plasma-renin in man". J. Physiol. 173: 408, 1964.

Brown, J. J., Davies, D. L., Lever, A. F. and Robertson, J. I. S.: "The assay of renin in single clomeruli in the normal rabbit and the appearance of the juxtaglomerular apparatus". J. Physiol. 176: 418, 1965.

Brown, J. J., Gleadle, R. I., Lawson, D. H., Lever, A. F., Linton, A. L., MacAdam, R. F., Prentice, E., Robertson, J. I. S. and Tree, M.: "Renin and acute renal failure: studies in man". Brit. Med. J. 1: 253, 1970.

Brun, C., Crome, C., Davidsen, H. G., Fabricius, J., Hansen, A. T., Lassen, N. A. and Munck, O.: "Renal blood flow in anuric human subject determined by use of radioactive Krypton 85". Proc. Soc. exp. Biol. (N. Y.) 89: 687, 1955.

Brun, C. and Munck, O.: "Lesions of the kidney in acute renal failure following shock". Lancet 1: 603, 1957.

Bull, G. M., Joekes, A. M. and Lowe, K. G.: "Renal function studies in acute tubular necrosis". Clin. Sci. 9: 379, 1950.

Bunag, R. D., Page, I. H. and McCubbin, J. W.: "Influence of dietary sodium on stimuli causing renin-release". Am. J. Physiol. 211: 1383, 1966a.

Bunag, R. D., Page, I. H. and McCubbin, J. W.: "Neural stimulation of release of renin". Circulation Res. 19: 851, 1966b.

Bunag, R. D., Page, I. H. and McCubbin, J. W.: "Inhibition of renin release by vasopressin and angiotensin". Cardiovas. Res. 1: 67, 1967.

Bunag, R. D., Vandor, A. J. and Kaneko, Y.: "Control of renin release". In: Renal Hypertension, Yearbook Med. Pub. 1968. Ed. Page, I. H. and McCubbin, J. A., Chap. 3, page 100.

Byrom, F. B.: "Morbid effects of vasopressin on the organs and vessels of rats". J. Path. Bact. 45: 1, 1937.

Byrom, F. B. : "Angiotensin and renal vascular damage". Brit. J. exp. Pathol. 45: 7, 1964.

Bywaters, E. G. L. and Beall, D. : "Crush injuries with impairment of renal function". Brit. Med. J. 1: 427, 1941.

Cameron, G. and Finckh, E. : "The production of acute hemolytic crisis by subcutaneous injection of glycerol". J. Path. Bact. 71: 165, 1956.

Carriere, S. : "Effect of norepinephrine, isoproterenol, and adrenergic blockers upon the intrarenal distribution of blood flow". Canad. J. Physiol. Pharmacol. 47: 199, 1969.

Carriere, S. , Daigneault, B. and Rochefort, F. : "Effect of retransfusion after hemorrhagic hypotension on intrarenal distribution of blood flow in dogs". J. Clin. Invest. 49: 2205, 1970.

Carriere, S. and Friborg, J. : "Intrarenal blood flow and PAH extraction during angiotensin infusion". Am. J. Physiol. 217: 1708, 1969.

Carriere, S. , Friborg, J. and Guay, J. P. : "Vasodilators, intrarenal blood flow, and natriuresis in the dog". Am. J. Physiol. 221: 92, 1971.

Carriere, S. , Thorburn, G. D. , O'Morchoe, C. C. C. and Barger, A. C. : "Intrarenal distribution of blood flow in dogs during hemorrhagic hypotension". Circulation Res. 19: 167, 1966.

Carroll, R. , Kovacs, K. and Tapp, E. : "The pathogenesis of glycerol-induced renal tubular necrosis". J. Path. Bact. 89: 573, 1965.

Collier, R. O. and Swann, H. G. : "Relation of kidney size to blood pressure". Am. J. Physiol. 220: 488, 1971.

Conn, H. L. Jr. , Wilds, L. and Helwig, J. : "A study of the renal circulation, tubular function and morphology, and urinary volume and composition in dogs following mercury poisoning and transfusion of human blood". J. Clin. Invest. 23: 732, 1954.

Cook, W. J. , Gordon, D. B. and Peart, W. S. : "The location of renin in the rabbit kidney". J. Physiol. 135: 46, 1957.

Cook, W. F. and Pickering, G. W. : "The location of renin in the rabbit kidney". J. Physiol. 149: 526, 1959.

Cosgrove, M. D. and Evans, K. : "The use of inert radioactive gases to measure blood flow in the perfused kidney". In: Blood Flow Through Organs and Tissues. Ed. Bain, W. H. and Harper, A. M., Livingstone, 1968, page 429.

Davies, D. J. : "The patterns of renal infarction caused by different types of temporary ischaemia". J. Pathol. 102: 151, 1970.

Davies, J. O. : "What signals the kidney to release renin?" Circulation Res. 28: 301, 1971.

Davis, B. B., Walter, M. J. and Murdaugh, H. V. Jr. : "Renal response to graded saline challenge". Am. J. Physiol. 217: 1604, 1969.

Day, M. D., McCubbin, J. W. and Page, I. H. : "Limited hypertensive effect of infusion of angiotensin". Am. J. Physiol. 209: 264, 1965.

De Champlain, J., Genest, J., Veyratt, R. and Boucher, R. : "Factors controlling renin in man". Archives of Int. Med. 117: 355, 1966.

De Wardener, H. : "Intrarenal pressure in experimental tubular necrosis". Lancet 1: 58, 1955.

DiBona, G. F., McDonald, F. D., Flamenbaum, W., Dammin, G. J. and Oken, D. E. : "Maintenance of renal function in salt loaded rats despite severe tubular necrosis induced by $HgCl_2$". Nephron. 8: 205, 1971.

Diomi, P., Matheson, N. A., Norman, J. N. and Shearer, J. R. : "Renal effects of dextran 40 during prolonged or repeated renal hypoperfusion". Surgery 130: 658, 1970.

DiSalvo, J. and Fell, C. : "Drug evoked neural responses in the canine renal vascular bed". Am. J. Physiol. 220: 1486, 1971.

Dobson, E. L. and Warner, G. F. : "Measurement of regional sodium turnover rates and their application to the estimation of regional blood flow". Am. J. Physiol. 189: 269, 1957.

Earley, L. E. and Friedler, R. M. : "Changes in renal blood flow and possibly the intrarenal distribution of blood during the natriuresis accompanying saline loading in the dog". J. Clin. Invest. 44: 929, 1965.

Elkin, M. and Meng, C. H. : "Angiographic study of the effect of vasopressors - epinephrine and levarterenol - on renal vascularity". Am. J. Roentgenol. 93: 904, 1965.

Elkin, M. and Meng, C. H. : "The effect of angiotensin on renal vascularity in dogs". Am. J. Roentgenol. 98: 927, 1966.

Epstein, M., Hollenberg, N. K. and Merrill, J. P. : "The pattern of the renal vascular response to epinephrine in man". Proc. Soc. exp. Biol. Med. 134: 720, 1970a.

Epstein, M., Berk, D. P., Hollenberg, N. K., Adams, D. F., Chalmers, T. C., Abrams, H. L. and Merrill, J. P. : "Renal failure in the patient with cirrhosis". Am. J. Med. 49: 175, 1970b.

Finckh, E. S. : "The indirect action of subcutaneous injections of glycerol on the renal tubules in the rat". J. Path. Bact. 78: 197, 1959.

Finckh, E. S. : "The pathogenesis of uraemia in acute renal failure". The Lancet II: 330, 1962.

Finckh, E. S., Jeremy, D. and Whyte, H. M. : "Structural renal damage and its relation to clinical features in acute oliguric renal failure". Quart. J. Med. New Series 31: 429, 1962a.

Fisher, R. D., Grünfeld, J. P. and Barger, A. C. : "Intrarenal distribution of blood flow in diabetes insipidus: role of ADH". Am. J. Physiol. 219: 1348, 1970.

Flamenbaum, W., McDonald, F. D., DiBona, G. F. and Oken, D. E. : "Micropuncture study of renal tubular factors in low dose mercury poisoning". Nephron. 8: 221, 1971.

Flamenbaum, W. and McNeil, J. : "Renal hemodynamics in uranyl nitrate induced acute renal failure". (Abst.) Am. Soc. Nephrol. 5th Annual Meeting, page 22, 1971.

Flanigan, W. J. and Oken, D. E. : "Renal micropuncture study of the development of anuria in the rat with mercury-induced acute renal failure". J. Clin. Invest. 44: 449, 1965.

Freed, T. A., Hager, H. and Vinik, M. : "Effects of intra-arterial acetylcholine on renal arteriography in normal humans". Am. J. Roentgenol., Rad. Therapy Nucl. Med. 104: 312, 1968.

Freed, T. A., Neal, M. P. and Vinik, M. : "The effect of bradykinin on renal arteriography". Am. J. Roentgenol. 102: 776, 1968.

Freed, T. A. and Vinik, M. : "Effect of acetylcholine on renal arteriography". Investig. Radiol. 3: 81, 1968.

Friedman, S. M., Johnson, R. L. and Friedman, C. L. : "The pattern of recovery of renal function following renal artery occlusion in the dog". Circulation Res. 2: 231, 1954.

Friedman, S. M. and Friedman, C. L. : "Effects of ions on vascular smooth muscle". In: Handbook of Physiology, Circulation, Vol. II: 1135, 1963.

Fung, H., McIvor, G. and Thomson, A. E. : "Effect of unilateral renal denervation and of salt loading on glycerol-induced acute renal failure in the rat". Clin. Res. 18: 746, 1970.

Gertz, K. H., Mangos, J. A., Braun, G. and Pagel, H. O. : "Pressure in the glomerular capillaries of the rat kidney and its relation to arterial blood pressure". Arch. Ges. Physiol. 288: 369, 1966.

Gill, J. R. and Bartter, F. C. : "Adrenergic nervous system in sodium metabolism. II. Effects of guanethidine on the renal response to sodium deprivation in normal man". New Engl. J. Med. 275: 1466, 1966.

Gill, J. R. Jr. and Casper, A. G. T. : "Role of the sympathetic nervous system in the renal response to hemorrhage". J. Clin. Invest. 48: 915, 1969.

Gill, J. R., Mason, D. T. and Bartter, F. C. : "Adrenergic nervous system in sodium metabolism: effects of guanethidine and sodium-retaining steroids in normal man". Part I. J. Clin. Invest. 43: 177, 1964.

Ginsburg, M. and Brown, L.M.: "Effects of anaesthetics and haemorrhage on the release of neurohypophyseal antidiuretic hormone". Brit. J. Pharmacol. 11: 236, 1956.

Goldberg, M.: "Studies of the acute renal effects of hemolyzed red blood cells in dogs including estimates of renal blood flow with krypton". J. Clin. Invest. 41: 2112, 1962.

Goormaghtigh, N.: "Vascular and circulatory changes in renal cortex in the anuric crush-syndrome". Proc. Soc. exp. Biol. (N. Y.) 59: 303, 1945.

Goormaghtigh, N.: "The renal anteriolar changes in the anuric crush syndrome". Am. J. Path. 23: 513, 1947.

Gosling, J. A.: "Observations on the distribution of intrarenal nervous tissue". Anat. Res. 163: 31, 1969.

Grandchamp, A., Ayer, G. Jr., Scherrer, J.R. and Truniger, B.: "Intrarenal hemodynamics of the rat kidney determined by the Xenon washout technique". Nephron. 8: 33, 1971.

Greenway, C. V. and Stark, R. D.: "Vascular responses of the spleen to rapid hemorrhage in the anaesthetized cat". J. Physiol. 204: 169, 1969.

Gross, F., Bock, K. D. and Turrian, H.: "Untersuchungen über die blutdruckwirkung von angiotensin". Helvet. physiol. et pharmacol. Acta 19: 42, 1961.

Gross, F., Schaechtelein, G., Brunner, H. and Peters, G.: "The role of the renin-angiotensin system in blood pressure regulation and kidney function". Canad. Med. Ass. J. 90: 258, 1964.

Gross, F., Brunner, H. and Ziegler, M.: "Renin-angiotensin system, aldosterone and sodium balance". Recent Prog. Hormone Res. 21: 119, 1965.

Gross, F., Khairallah, P. A., McGiff, J. C. and Bunag, R. D.: "Pharmacology of angiotensin". In: Renal Hypertension. Ed. Page, I. H. and McCubbin, J. W. Yearbook Med. Pub. 1968, Chap. 5, page 135.

Grünfield, J. P., Raphaël, J. C., Bankir, L., Kleinknecht, D. and Barger, A. C.: "Intrarenal distribution of blood flow". Adv. in Nephrol. 1: 125, 1971.

Haber, E., Page, L. B. and Richards, F. F.: "Radioimmunoassay employing gel filtration". An. Biochem. 12: 163, 1965.

Hamilton, P. B., Phillips, R. A. and Hiller, A.: "Duration of renal ischemia required to produce uremia". Am. J. Physiol. 152: 517, 1948.

Handley, C. A. and Moyer, J. H.: "Unilateral renal adrenergic blockade and the renal response to vasopressor agents and to hemorrhage". J. Pharmac. exp. Ther. 112: 1, 1954.

Harrison, H. E., Bunting, H., Ordway, N. K. and Albrink, W. S.: "The pathogenesis of the renal injury produced in the dog by hemoglobin or methemoglobin". J. exp. Med. 86: 339, 1947.

Hartroft, P. N.: "Histological and functional aspects of juxtaglomerular cells". In: Angiotensin Systems and Experimental Renal Diseases. Ed. Metcoff, J., Little, Brown & Co., Boston, 1963.

Harvey, R. B.: "Effects of acetylcholine infused into the renal artery of dogs". Am. J. Physiol. 211: 487, 1966.

Hatch, F. E. and Johnson, J. G.: "Intrarenal blood flow". Ann. Rev. Med. 20: 395, 1969.

Hatcher, C. R. J., Gagnon, J. A., Clarke, R. W. and Geever, E. F.: "Acute reversible and irreversible renal insufficiency in the dog". J. S. R. II: 36, 1962.

Hayes, J. M., Boonshaft, B., Maher, J. R., O'Connell, J. M. B. and Schreiner, G. E.: "Resistance to glycerol-induced hemoglobinuric acute renal failure". Nephron. 7: 155, 1960.

Henry, L. N., Lane, C. E. and Kashgarian, M.: "Micropuncture studies of pathophysiology of acute renal failure in the rat". Lab. Invest. 19: 309, 1968.

Herd, J. A. and Barger, A. C.: "Simplified technique for chronic catheterization of blood vessels". J. Appl. Physiol. 19: 791, 1964.

Hinshaw, L. B.: "Autoregulation in normal and pathological states including shock and ischemia". Circulation Res. 28: 1, 1971.

Hinshaw, L. B., Bradley, G.M. and Carlson, C.H.: "Effect of endotoxin on renal function in the dog". Am. J. Physiol. 196: 1127, 1959.

Hollenberg, N.K.: "The role of the sympathetic nervous system in the development and decompensation during hemorrhagic shock". Ph. D. Thesis, 1965. University of Manitoba.

Hollenberg, N.K., Epstein, M., Rosen, S.M., Basch, R.I., Oken, D.E. and Merrill, J.P.: "Acute oliguric renal failure in man: evidence for preferential renal cortical ischemia". Med. 47: 455, 1968.

Hollenberg, N.K., Epstein, M., Rosen, S.M. and Dammin, G.J.: "Vascular lesions of the transplanted human kidney - morphologic and hemodynamic studies in chronic rejection". Trans. Assoc. Amer. Physicians, 81: 274, 1968a.

Hollenberg, N.K., Retik, A.B., Rosen, S.M., Murray, J.E. and Merrill, J.P.: "The role of vasoconstriction in the ischemia of renal allograft rejection". Transplantation 6: 59, 1968b.

Hollenberg, N.K., Rosen, S.M., O'Connor, J.F., Potchen, E.J., Basch, R., Deally, J.B. Jr. and Merrill, J.P.: "Effect of aortography on renal hemodynamics in normal man". Investig. Radiol. 3: 92, 1968c.

Hollenberg, N.K., Adams, D.F., Oken, D.E., Abrams, H.L. and Merrill, J.P.: "Acute renal failure due to nephrotoxins renal hemodynamics and angiographic studies in man". New Engl. J.Med. 282, 1329, 1970a.

Hollenberg, N.K., Epstein, M., Guttmann, R.D., Conroy, M., Basch, R.I. and Merrill, J.P.: "Effect of sodium balance on intrarenal distribution of blood flow in normal man". J. Appl. Physiol. 28: 312, 1970b.

Hollenberg, N.K. and Merrill, J.P.: "Intrarenal perfusion in the young "essential" hypertensive: A subpopulation resistant to sodium restriction". Trans. Assoc. Am. Physicians, 83: 93, 1970.

Hollenberg, N.K., Adams, D.F., Rashid, A., Epstein, M., Abrams, H.L. and Merrill, J.P.: "Renal vascular response to salt restriction in normal man. Evidence against adrenergic mediation". Circulation 43: 845, 1971.

Houck, C.R.: "Alterations in renal hemodynamics and function in separate kidneys during stimulation of renal artery nerves in dogs". Am. J. Physiol. 167: 523, 1951.

Ingvar, D.H. and Lassen, N.A.: "Regional blood flow of the cerebral cortex determined by Krypton 85". Acta Physiol. Scand. 54: 325, 1962.

Innes, I.R. and Nickerson, M.: "Drugs acting on post-glanglionic adrenergic nerves endings and structures innervated by them (Sympathomimetic drugs)". In: The Pharmacological Basis of Therapeutics, Fourth Edition. Ed. Goodman, L.S. and Gilman, A., MacMillan, 1970, Chap. 24, page 478.

Jaenike, J.R.: "The renal lesion associated with hemoglobinemia: A study of the pathogenesis of the excretory defect in the rat". J. Clin. Invest. 46: 378, 1967.

Jirka, J., Ganz, V., Fencl, V., Cort, J.H. and Travicek, R.: "Measurement of renal blood flow in the intact kidney by local thermodilution during haemorrhagic hypotensin". Lancet 2: 692, 1961.

Johnston, H.H., Herzog, J.P. and Lauler, D.P.: "Effect of prostaglandin E_1 on renal hemodynamics, sodium and water excretion". Am. J. Physiol. 213: 939, 1967.

Kamm, D.E. and Levinsky, N.G.: "The mechanism of denervation natriuresis". J. Clin. Invest. 44: 93, 1965.

Katz, M.A., Blantz, R.C., Rector, F.C. and Seldin, D.W.: "Measurement of intrarenal blood flow. I. Analysis of microsphere method". Am. J. Physiol. 220: 1903, 1971.

Kety, S.S.: "The theory and applications of the exchange of inert gas at the lungs and tissues". Pharmacol. Rev. 3: 1, 1951.

Kew, M. C., Varma, R. R., Williams, H. S., Brunt, P. W., Hourigan, K. J. and Sherlock, S.: "Renal and intrarenal blood flow in cirrhosis of the liver". The Lancet 2: 504, 1971.

Kiley, J. E., Power, S. R. and Beebe, R. T.: "Acute renal failure. Eighty cases of renal tubular necrosis". New Engl. J. Med. 262: 481, 1960.

Kohot, F. and Kuska, J.: "Plasma renin activity in acute renal insufficiency". Nephron. 6: 115, 1969.

Koletsky, S.: "Effects of temporary interruption of renal circulation in rats". A. M. A. Arch. Path. 58: 592, 1954.

Korner, R. I.: "Effect of low oxygen and of carbon monoxide on the renal circulation in unanesthetized rabbits". Circulation Res. 12: 361, 1963.

Kramer, K., Thurau, K. and Deetjen, P.: "Hamodynamik des nierenmarks". Pflugers Arch. ges. Physiol. 270: 251, 1960.

Kubicek, W. G., Kottke, F. J., Laker, D. J. and Visscher, M. B.: "Renal function during arterial hypertension produced by chronic splanchnic nerve stimulation in the dog". Am. J. Physiol. 174: 397, 1953.

Ladefoged, J.: "Measurements of the renal blood flow in man with the $Xenon^{133}$ washout technique - A description of the method". Scand. J. Clin. Lab. Invest. 18: 299, 1966.

Ladefoged, J. and Pedersen, F.: "Renal blood flow in isolated kidneys measured with an electromagnetic flowmeter and by $Xenon^{133}$ and $Krypton^{85}$ washout techiques". Pflugers Arch. 299: 30, 1968.

Ladefoged, J. and Winkler, K.: "Hemodynamics in acute renal failure". Scand. J. Clin. Lab. Invest. 26: 83, 1970.

Lambert, P. P., Kahn, R. J., Gottignies, P., Vanherweghem, J. L. et Frafchamps, R.: "Measurement of renal blood flow by the local thermodilution method in the renal artery". Nephron. 8: 125, 1971.

Langston, J. B., Guyton, A. C. and Gillespie, W. J. Jr.: "Acute effect of changes in renal arterial pressure and sympathetic blockade on kidney function". Am. J. Physiol. 147: 595, 1959.

Langston, J. B., Guyton, A. C., DePoyster, J. H. and Armstrong, G. G. Jr.: "Changes in renal function resulting from norepinephrine infusion". Am. J. Physiol. 202: 893, 1962.

Lauson, H. D., Bradley, S. E. and Cournand, A.: "The renal circulation in shock". J. Clin. Invest. 23: 381, 1944.

Lavender, J. P. and Sherwood, T.: "The renal cortex angiogram and renal blood flow: Studies of renal cortical perfusion under differing functional states". Brit. J. Radiol. 44: 505, 1971.

Lee, J. B., Covino, B. G., Takman, B. H. and Smith, E. R.: "Renomedullary vasodepressor substance, medullin: isolation, chemical characterization and physiological properties". Circulation Res. 17: 57, 1965.

Lee, P. and Lickett, M. F.: "Some actions and isoprenaline and orciprenaline on perfused cat kidneys". Brit. J. Pharmacol. 25: 152, 1963.

Lees, P. and Pockett, M. F.: "A study of the beta-receptors in the rat kidneys". Brit. J. Pharmacol. 20: 135, 1963.

Lewis, D. H. and Bergentz, Sven-erik: "Renal blood flow measurement with Xenon^{133} at the time of operation for renal artery stenosis". Surgery 59: 1043, 1966.

Lewis, D. H., Bergentz, S. E., Brunius, V., Ekman, H., Gelin, L. E. and Hood, B.: "Blood flow in kidney transplants". Scand. J. Urol. Nephrol. 2: 36, 1968.

Lewis, D. H. and Fritjofsson, A.: "Comparison of Xenon^{133} washout curves from the kidney with direct measurement of renal venous outflow". Scand. J. Urol. Nephrol. 2: 62, 1968.

Liebau, H., Distler, A. and Wolff, H. P.: "Untersuchungen zur indireckten sympathomimetischen Wirkung von Angiotensin au isolierten Blut gefässen". Klin. Wchnschr. 44: 322, 1966.

Logan, A., Jose, P., Eisner, G., Lilienfield, L. and Slotkoff, L.: "Intracortical distribution of renal blood flow in hemorrhagic shock in dogs". Circulation Res. 29: 257, 1971.

Lucke, B.: "Lower nephron nephrosis". Mil. Surgeon, 99: 371, 1946.

Ludin, H., Elke, M., Fehr, H. and Thoelen, H.: "Correlation of renal size, renal artery calibre and effective renal plasma flow in man". Acta Radiol. Diag. 6: 296, 1967.

Macfarlane, D.M.: "A renorenal vasoconstrictor reflex induced by acetylcholine". Am. J. Physiol. 218: 851, 1970.

McDonald, S.J. and de Wardener, H.E.: "Some observations on the production of a hypo-osmotic urine during the administration of 0.9% saline and vasopressin in the dog". Clin. Sci. 28: 445, 1965.

McDonald, F.D., Thiel, G., Wilson, D.R., DiBona, G.F. and Oken, D.E.: "The prevention of acute renal failure in the rat by long-term saline loading: A possible role of the renin-angiotensin axis". Proc. Soc. exp. Biol. Med. 131: 610, 1969.

McGiff, J.C. and Burns, B.P.: "Role of acetylcholine in the renal vasoconstrictor response to sympathetic nerve stimulation in the dog". Circulation Res. XX: 616, 1967.

McGiff, J.C. and Fasy, T.M.: "The relationship of renal vascular activity of angiotensin II to the autonomic nervous system". J. Clin. Invest. 44: 1911, 1965.

McGiff, J.C., Crowshaw, K., Terragno, N.A., Lonigro, A.J., Strand, J.C., Williamson, M.A., Lee, J.B. and Ng, K.K.F.: "Prostaglandin-like substances appearing in canine renal venous blood during renal ischemia". Circulation Res. 27: 765, 1970.

McGiff, J.C. and Itskovitz, H.O.: "Loss of the renal vasoconstrictor activity of angiotensin II during renal ischemia". J. Clin. Invest. 43: 2359, 1964.

McKay, D.G., Latour, J.G. and Parrish, M.H.: "Activation of hageman factor by alpha-adrenergic stimulation". Thrombos. Diathes. haemorh. 23: 417, 1970.

McKenna, O.C. and Angelakos, E.T.: "Adrenergic innervation of the canine kidney". Circulation Res. 22, 345, 1968.

McKenzie, I. F. C. and Whittingham, S.: "Deposits of immunoglobulin and fibrin in human allografted kidneys". Lancet II: 1313, 1968.

McKenzie, J. K., Ryan, J. W. and Lee, M. R.: "Effect of laparotomy on plasma renin activity in the rabbit". Nature 215: 542, 1967.

McKenzie, J. K.: Unpublished observations, 1971.

McLean, D. and Thomson, A. E.: "Effect of phenoxybenzamine on glycerol-induced acute renal failure in the rat". Fed. Proc. 29: 1313, 1970.

McNay, J. L. and Abe, Y.: "Redistribution of cortical blood flow during renal vasodilatation in dogs". Circulation Res. 27: 1023, 1970.

McNeill, J. R., Stark, R. D. and Greenway, C. V.: "Intestinal vasoconstriction after hemorrhage: roles of vasopressin and angiotensin". Am. J. Physiol. 219: 1342, 1970.

Mailloux, L., Swartz, C. D., Capizzi, R., Kim, K. E., Onesti, G., Ramirez, O. and Brest, A. N.: "Acute renal failure after low molecular weight dextran administration". New Engl. J. Med. 277: 1113, 1967.

Mallory, T. B.: "Hemoglobinuric nephrosis in traumatic shock". Am. J. Clin. Path. 17: 427, 1947.

Maluf, N. S. R.: "Factors inducing renal shut-down from lysed erythrocytes: an experimental study". Ann. Surg. 130: 49, 1949.

Mancini, P. and Alexxandro, P.: "A computer program for multi-exponential filting by the peeling method". Computers and Biomed. Res. 3: 1, 1970.

Mandel, M. and Sapirstein, L.: "Effect of angiotensin infusion on renal blood flow and regional vascular resistance in the rat". Circulation Res. 10: 807, 1962.

Martinez-Maldonado, M., Eknoyan, G. and Suki, W. N.: "Natriuretic effects of vasopressin and cyclic AMP: possible site of action in the nephron". Am. J. Physiol. 220: 2013, 1971.

Mason, A. D. Jr., Alexander, J.W. and Teschan, P. E.: "Studies in acute renal failure. I. Development of a reproducible lesion in experimental animals". J. surg. Res. 3: 430, 1963a.

Mason, A. D. Jr., Teschan, P. E. and Muirhead, E. E.: "Studies in acute renal failure. III. Renal histologic alterations in acute renal failure in the rat". J. surg. Res. 3: 450, 1963b.

Massani, Z. M., Finkielman, S., Worcel, M., Agrest, A. and Paladini, A. C.: "Angiotensin blood levels in hypertensive and non hypertensive diseases". Clin. Sci. 30: 473, 1966.

Menefee, M. G., Mueller, C. B., Miller, T. B., Myers, J. K. and Bell, A. L.: "Experimental studies in acute renal failure. II. Fine structure changes in tubules associated with renal failure induced by globin". J. exp. Med. 120: 1139, 1964.

Merrill, J. P.: "Kidney diseases: acute renal failure". Ann. Rev. Med. 11: 127, 1960.

Merrill, J. P.: "Acute renal failure". In: Diseases of the Kidney. Ed. Strauss, M. B. and Welt, L. G., Little, Brown & Co., Second Edition, 1971. Chap. 17, page 637.

Miller, J. H. and McDonald, R. K.: "The effect of hemoglobin on renal function in the human". J. Clin. Invest. 30: 1033, 1951.

Mogil, R. A., Itskovitz, H. D., Russell, J. H. and Murphy, J. J.: "Renal innervation and renin activity in salt metabolism and hypertension". Am. J. Physiol. 216: 693, 1969.

Montague, F. E. and Wilson, F. L. Jr.: "Effect of epinephrine and shock and PAH extraction by the rabbit kidney". Am. J. Physiol. 159: 581, 1949.

Moses, J. B.: "The distribution of renal ischemia produced by epinephrine and norepinephrine as demonstrated by the fluorescent dye vasoflavine". J. Urol. 68: 558, 1952.

Moss, A. J. and Schenk, E. A.: "Cardiovascular effects of sustained norepinephrine infusions in dogs". Circulation Res. 27: 1013, 1970.

Moyer, J. H. and Handley, C. A.: "Norepinephrine and epinephrine effect on renal hemodynamics". Circulation 5: 91, 1952.

Muehrcke, R. C.: "Acute Renal Failure - Diagnosis and Managements". C. V. Mosby, 1969.

Mueller, C. B.: "The pathogenesis of acute renal failure". In: Renal Failure. Ed. Brest, A. N. and Moyer, J. H., Lippincott Press, 1967, Part V, page 245.

Muirhead, E. E., Brown, G. B., Germain, G. S. and Leach, B. E.: "The renal medulla as an antihypertensive organ". J. Lab. Clin. Med. 76: 641, 1970.

Müller-Berghaus, G., Davidson, E. and McKay, D. G.: "Prevention of the generalized Shwartzman reaction in pregnant rats by alpha-adrenergic blockade". Obstet. and Gynec. 30: 774, 1967.

Müller-Berghaus, G. and McKay, D. G.: "Prevention of the generalized Shwartzman reaction in pregnant rats by alpha-adrenergic blocking agents". Lab. Invest. 17: 276, 1967.

Munck, O., de Bono, E. and Mills, I. A.: "Distribution of blood flow in the renal cortex during saline loading". Clin. Sci. 38: 699, 1970.

Myburgh, J. A., Cohen, I., Gecelter, L., Meyers, A. M., Abrahams, C., Furman, K. I., Goldberg, B. and Van Blerk, P. J. P.: "Hyperacute rejection in human kidney allografts - Schwartzman or Arthus reaction?" New Engl. J. Med. 281: 131, 1969.

Myers, J. K., Miller, T. B., Mueller, C. B. and Storrs, D.: "The role of ADH in experimental acute renal failure". (Abst). Fed. Proc. 12: 661, 1963.

Newsome, H. H. Jr., Kafka, M. S. and Bartter, F. C.: "Intrarenal blood flow in dogs with constriction of the inferior thoracic vena cava". Am. J. Physiol. 221: 48, 1971.

Ng, K. K. F. and Vane, J. R.: "Fate of angiotensin I in the circulation". Nature 218: 144, 1968.

Nickerson, M.: "The pharmacology of adrenergic blockade". Pharmacol. Rev. 1: 27, 1949.

Nickerson, M.: "Vascular adjustments during the development of shock". C. M. A. Journ. 103: 853, 1970.

Nickerson, M. and Hollenberg, N.K.: "Blockade of alpha-adrenergic receptors". In: Physiological Pharmacology, Vol. 4, Part D, Autonomic Nervous System Drugs. Ed. Root, W.S. and Hofmann, F.G., Academica Press, 1967, page 243.

Nickerson, M. and Sutter, M.C.: "Angiotensin in shock". Canad. Med. Ass. J. 90: 325, 1964.

Norman, J.N., Irvin, T.T. and Smith, G.: "The effect of inspired oxygen tension on renal blood flow". In: Blood Flow Through Organs and Tissues. Ed. Bain, W.H. and Harper, A.M., Livingstone, 1968, p. 449.

Ochoa, E., Finkihman, S. and Agrest, A.: "Angiotensin blood levels during evolution of acute renal failure". Clin. Sci. 38: 225, 1970.

Oken, D.E., Arce, M.L. and Wilson, D.R.: "Glycerol-induced hemoglobinuric acute renal failure in the rat. I. Micropuncture study of the development of oliguria". J. Clin. Invest. 45: 724, 1966.

Oken, D.E., DiBona, G.F. and McDonald, F.D.: "Micropuncture studies of the recovery phase of myohemoglobinuric acute renal failure in the rat". J. Clin. Invest. 49: 730, 1970.

Oliver, J., McDowell, M. and Tracy, A.: "The pathogenesis of acute renal failure associated with traumatic tonic injury. Renal ischemia, nephrotoxic injury, and the ischemic episode". J. Clin. Invest. 30: 1305, 1951.

Olsen, J.S. and Skjoldborg, H.: "The fine structure of the renal glomerulus in acute anuria". Acta Path..et Microbiol. Scand. 70: 205, 1967.

Owen, K., Desautels, R. and Walter, C.W.: "Experimental renal tubular necrosis - the effect of pitresin". Surg. Forum 4: 459, 1953.

Penner, A. and Bernheim, A.I.: "Acute ischemic necrosis of the kidney". Archives Path. 30: 465, 1940.

Peter, G. and Grunner, H.: "Mannitol diuresis in hemorrhagic hypotension". Am. J. Physiol. 204: 555, 1963.

Pickering, G.: In: High Blood Pressure. Grune & Stratton Inc., N. Y., 1968.

Pilkington, L. A., Binder, R., de Haas, J. C. M. and Pitts, R. F.: "Intrarenal distribution of blood flow". Am. J. Physiol. 208: 1107, 1065.

Phillips, R. A., Dole, V. P., Hamilton, P. B., Emerson, K. Jr., Archibald, R. M. and Van Slyke, D. D.: Effects of acute hemorrhagic and traumatic shock on renal function of dogs". Am. J. Physiol. 145: 314, 1945.

Phillips, R. A. and Hamilton, P. B.: "Effect of 20, 60 and 120 minutes of renal ischemia on glomerular and tubular function". Am. J. Physiol. 152: 523, 1948.

Pomeranz, B. H., Birtch, A. G. and Barger, A. C.: "Neural control of intrarenal blood flow". Am. J. Physiol. 215: 1067, 1968.

Rashid, A., Hollenberg, N. K. and Merrill, J. P.: "The effect of intrarenal infusion of acetylcholine on the renal blood flow in normal man". Clin. Res. 18: 749, 1970.

Reubi, F. C. and Schroeder, H. A.: "Can vascular shunting be induced in the kidney by vasoactive drugs?" J. Clin. Invest. 28: 114, 1949.

Reubi, F. C., Grosswerler, N. and Gürtler, R.: "Renal circulation in man studied by means of a dye-dilution method". Circulation 33: 426, 1966.

Retik, A. B., Hollenberg, N. K., Rosen, S. M., Harrison, J. H., Merrill, J. P. and Murray, J. E.: "Hemodynamic changes following renal transplantation". Trans. Am. Assoc. Genito-Urin. Surgeons 59: 56, 1967.

Retik, A. B., Hollenberg, N. K., Rosen, S. M., Merrill, J. P. and Murray, J. E.: "Cortical ischemia in renal allograft rejection". Surg. Gynecol. Obstet. 124: 989, 1967.

Retik, A. B., Rosen, S. M., Hollenberg, N. K., Murray, J. E. and Harrison, J. H.: "Intrarenal distribution of blood flow in canine renal allografts treated with immunosuppressive drugs". J. Urol. 101: 482, 1969.

Riggs, D. S. : "The mathematical approach to physiological problems: a critical primer". Williams and Wilkins Co., Baltimore, 1963.

Romero, J. C., Staneloni, R. J., Dufau, M. L., Dohmen, R., Binia, A., Kliman, B. and Fasciolo, J. C. : "Changes in fluid compartments, renal hemodynamics, plasma renin and aldosterone secretion induced by low sodium intake". Metabolism 17: 10, 1968.

Rosen, S. M., Hollenberg, N. K., Dealy, J. B. Jr. and Merrill, J. P. : "Measurement of the distribution of blood flow in the human kidney using the intra-arterial injection of Xenon^{133}. Relationship to function in the normal and transplanted kidney". Clin. Sci. 34: 287, 1968.

Ruiz-Guinazu, A. : "Alterations of the glomerular filtration rate in acute renal failure". In: Pathogenesis and Clinical Findings With Renal Failure. Ed. Gessler, Schroder and Weidinger, G. T. Verlag of Stuttgart, 1971, page 23.

Ruiz-Guinazu, A., Coelho, J. B. and Paz, R. A. : "Methemoglobin-induced acute renal failure in the rat". Nephron. 4: 257, 1967.

Sartorius, O. W. and Burlington, H. : "Acute effects of denervation on kidney function in the dog". Am. J. Physiol. 185: 407, 1956.

Schnermann, J., Nagel, W. and Thurau, K. : "Die frühdistale Natriumkonzentration in Rattenniern nach renaler Ischämie und hämorrhagischer Hypotension". Pflugers Arch. ges. Physiol. 287: 296, 1966.

Schrier, R. W., McDonald, K. M., Jagger, P. I. and Lauler, D. P. : "The role of the adrenergic nervous system in the renal response to acute extracellular fluid volume expansion". Proc. Soc. exp. Biol. Med. 125: 1157, 1967.

Selkurt, E. E. : "The renal circulation". Handbook of Physiology - Circulation II, Chap. 43, 1963.

Selkurt, E. E. : "Renal blood flow and renal clearance during hemorrhagic shock". Am. J. Physiol. 145: 699, 1946.

Sevitt, S. : 'Pathogenesis of traumatic uraemia". Lancet 2: 135, 1959.

Shanbour, L. L., Lindeman, R. D., Archer, L. T., Tung, S. H. and Inshaw, L. B.: "Improvement of renal hemodynamics in endotoxin shock with dopamine, phenoxybenzamine and dextran". J. Pharmac. exp. Ther. 176: 383, 1971.

Silverblatt, F., Turck, M. and Bulger, R.: "Nephrotoxicity due to cephaloridine: a light and electron microscopic study in rabbits". J. Inf. Dis. 122: 33, 1970.

Sims, E., Golberg, I., Kelley, J. and Sisco, B.: "Glomerular perfusion during acute renal insufficiency from mercury poisoning in the rat". J. Lab. Clin. Med. 54: 44, 1959.

Skinner, S. L., McCubbin, J. W. and Page, I. H.: "Renal baroceptor control of renin secretion". Science 141: 814, 1963.

Smith, H. W., Rovenstine, E. A., Goldring, W., Chasis, H. and Ranges, H. A.: "The effects of spinal anesthesia on the circulation in normal, unoperated man with reference to the autonomy of the arterioles, and especially those of the renal circulation". J. Clin. Invest. 18: 319, 1939.

Solomon, H. S., Hollenberg, N. K. and Merrill, J. P.: "Renal hemodynamics in acute renal failure: A comparison of Xenon washout and microsphere distribution methods in the dog". Am. Soc. Nephrol. 5th Annual Meeting (Abst.), page 75, 1971.

Spencer, M. P., Denison, A. B. Jr. and Green, H. D.: "The direct renal vascular effects of epinephrine and norepinephrine before and after adrenergic blockade". Circulation Res. 2: 537, 1954.

Spinazzola, A. J. and Sherrod, T. R.: "The effects of serotonin (5-hydroxytryptamine) on renal hemodynamics". J. Pharmac. exp. Ther. 119: 114, 1957.

Steinhausen, M., Eisenbach, G. M. and Helmstadter, V.: "Concentration of lissamine green in proximal tubules of antidiuretic and mercury poisoned rats and the permeability of those tubules". Pflügers Arch. ges. Physiol. 311: 1, 1969.

Stiehm, E. R. and Trygstad, C. W.: "Split products of fibrin in human renal disease". Am. J. Med. 46: 774, 1969.

Stinson, J. M., Barnes, A. B., Zakheim, R. M., Chimoskey, J. E., Spinelli, F. R. and Barger, A. C.: "Reflex cholinergic vasodilatation during renal artery constriction in the unanaesthetized dog". Am. J. Physiol. 217: 239, 1969.

Sundsfjord, J. A. : "Renal production of angiotensin II". Lancet 2: 807, 1969.

Sutter, M. C. : "Noradrenaline - A study of its role in production of and protection against shock". Ph. D. Thesis, 1963, University of Manitiba.

Sweet, C. S. , Kadowitz, P. J. and Brody, M. J. : "Another humoral substance that enhances adrenergic responsiveness during acute renal ischemia". Nature 231: 263, 1971.

Tagawa, H. , Vander, A. J. , Bonjour, J. P. and Malvin, R. L. : "Inhibition of renin secretion by vasopressin in unanesthetized sodium-deprived dogs". Am. J. Physiol. 220: 949, 1971.

Takeuchi, J. , Ishikawa, I. , Inasaka, T. , Sakai, S. , Shinoda, A. , Takada, A. and Homada, N. : "Intrarenal distribution or blood flow in man: A new analytic method for dye-dilution curves". Circulation 42: 347, 1970.

Thiel, G. , McDonald, F. D. and Oken, D. E. : "Micropuncture studies of the basis for protection of renin depleted rats from glycerol-induced acute renal failure". Nephron. 7: 67, 1970.

Thiel, G. , Wilson, D. R. , Arce, M. L. and Oken, D. E. : "Glycerol-induced hemoglobinuric acute renal failure in the rat. II. The experimental model, predisposing factors, and pathophysiologic features". Nephron. 4: 276, 1967.

Thomson, A. E. : "Adrenergic blockade in clinical shock of mixed etiology". In: Shock and Hypotension. Ed. Mills, L. C. and Moyer, J. H. , Grune and Stratton, 1965, page 478.

Thorburn, G. D. , Kopald, H. H. , Herd, J. A. , Hollenberg, M. , O'Marchoe, C. C. and Barger, A. C. : "Intrarenal distribution of nutrient blood flow determined with Krypton-85 in the unanesthetized dog". Circulation Res. 13: 290, 1963.

Thurau, K. and Levine, D. Z. : "The Renal Circulation". In: The Kidney. Ed. Rouiller, C. and Muller, A. F. , Academic Press, 1971, Vol. III, page 1.

Trueta, J., Barclay, A.E., Daniel, P.M., Franklin, K.J. and Prichard, M.M.L.: "Studies of the Renal Circulation". Blackwell Scientific Publications, Oxford, 1947.

Truniger, B., Rosen, S.M., Kriek, H., Merrill, J.P. and Murray, J.E.: "Intrarenal distribution or blood flow in the rejecting homotransplanted dog kidney". Surg. Forum 16: 254, 1965.

Tu, W.H.: "Plasma renin activity in acute tubular necrosis and other renal diseases associated with hypertension". Circulation 31: 686, 1965.

Ueda, H., Tagawa, H., Ishii, M. and Kaneko, Y.: "Effect of renal denervation on release and content of renin in anesthetized dogs". Jap. Heart J. 8: 156, 1967.

Vander, A.J.: "Effect of acetylcholine, atropine and physostigmine on renal function in the dog". Am. J. Physiol. 206: 492, 1964.

Vander, A.J.: "Effect of catecholamines and the renal nerves on renin secretion in anesthetized dogs". Am. J. Physiol. 209: 659, 1965.

Vander, A.J. and Geelhoed, G.W.: "Inhibition of renin secretion by angiotensin II". Proc. Soc. exp. Biol. Med. 120: 399, 1965.

Vander, A.J.: "Direct effects of prostaglandin on renal function and renin release in anesthetized dog". Am. J. Physiol. 214: 218, 1968.

Vasalli, P. and McCluskey, R.T.: "The coagulation process and glomerular disease". Am. J. Med. 39: 179, 1965.

Vaughn, D.L., Bersentes, T., Kirschbaum, T.H. and Assali, N.S.: "Hemeostatic role of the renal-adrenal axis in bacteremic shock". Am. J. Obst. Gynecol. 99: 208, 1967.

Villareal, H., Nieto de Pascual, H., Arcila, H., Lopez Vinas, C. and Pierra, P.: "The action of angiotensin on renal transport of sodium in the dog". Nephron. 1: 338, 1964.

Wakim, K.G., Herrick, J.F., Baldes, E.J. and Mann, F.C.: "The effect of pitressin on renal circulation and urine secretion". J. Lab. Clin. Med. 27: 1013, 1942.

Walker, J. G., Silva, H., Lawson, T. R., Ryder, J. A. and Shaldon, S.: "Renal blood flow in acute renal failure measured by renal arterial infusion of indocyanine green". Proc. Soc. exp. Biol. Med. 112: 932, 1963.

Wallin, J. D., Blantz, R. C., Katz, M. A., Andreucci, V. E., Rector, F. C. Jr. and Seldin, D. W.: "Effect of saline diuresis on intrarenal blood in the rat". Am. J. Physiol. 221: 1297, 1971.

Wallin, J. D., Rector, F. C. Jr. and Seldin, D. W.: "Measurement of intrarenal plasma flow with antiglomerular basement-membrane antibody". Am. J. Physiol. 221: 1621, 1971.

Waugh, D. and Pearl, M. J.: "Serotonin-induced acute nephrosis and renal cortical necrosis in rats". Am. J. Path. 36: 431, 1960.

Weidmann, P., Maxwell, M. H., Lupu, A. N., Lewin, A. J. and Massry, S. G.: "Plasma renin activity and blood pressure in terminal renal failure". New Engl. J. Med. 285: 757, 1971.

Weitsen, H. A. and Norvell, J. E.: "Cholinergic innervation of the auto-transplanted canine kidney". Circulation Res. 25: 535, 1969.

Willis, L. R., Ludens, J. H., Hook, J. B. and Williamson, H. E.: "Mechanism of natriuretic action of bradykinin". Am. J. Physiol. 217: 1, 1969.

Willis, L. R., Ludens, J. H. and Williamson, H. E.: "Dependence of the natriuretic action of acetylcholine upon an increase in renal blood flow". Proc. Soc. exp. Biol. Med. 128: 1069, 1968.

Wilson, D. R., Thiel, G., Arce, M. L. and Oken, D. E.: "The role of concentrating mechanism in the development of acute renal failure: micropuncture studies using diabetes insipidus rats". Nephron. 6: 128, 1969.

Wilson, D. R., Thiel, G., Arce, M. L. and Oken, D. E.: "Glycerol-induced hemoglobinuric acute renal failure in the rat. III. Micropuncture study of the effects of mannitol and isotonic saline on individual nephron function". Nephron. 4: 337, 1967.

Zierler, K. L. : "Equations for measuring blood flow by external monitoring of radioisotopes". Circulation Res. 16: 309, 1965.

Zimmerman, B. C. : "Effect of acute sympathectomy on responses to angiotensin and noradrenaline". Circulation Res. 11: 780, 1962.

Zimmerman, B. C., Abbound, L. M. and Eckstein, J. W. : "Effect of norepinephrine and angiotensin on total and venous resistance in the kidney". Am. J. Physiol. 206: 701, 1964.

SECTION VII

APPENDIX

CASE SUMMARIES

Mr. G. L.
WGH 203635

This 78 year old white male was admitted to the Winnipeg General Hospital on 3rd Dec., 1971 with a three-week history of intermittent abdominal pain. On evening of admission, patient vomited large amounts of foul smelling dark fluid following a severe bout of lower abdominal pain after having eaten supper. Patient denied melena, bloody stools and hematemesis. There was no history of peptic ulcer. He had arthritis for over 30 years and this had been treated with Entrophen^{R} and Darvon^{R}.

Physical examination and subsequent investigations on day of admission revealed generalized abdominal tenderness and guarding. Hemoglobin was 8.5 gms%; WBC 9.000; with a B.P. 140/89; regular pulse 124/min. BUN on admission was 29 mg%. Urine sediment was essentially negative. There was free air under the right diaphragm. Laparotomy was performed on the same day. There was about 600 ml of purulent peritoneal fluid present. The transverse colon was dilated. The descending and sigmoid colon was grossly edematous and acutely inflamed. A diagnosis of perforated sigmoid colon secondary to diverticulitis was made and a transverse colostomy was performed. Parenteral Kanamycin and Ampicillin was started

Post-operatively, the patient improved to the point of taking

oral fluids. On 7th Dec., 1971, 4 days post-operatively, the patient developed sudden chills, tachycardia and hyperventilation. On 8th Dec., 1971, he became very restless, confused, hypotensive, oliguric and developed a pleural rub. The diagnosis of pulmonary embolism was made and was confirmed by lung scan. Heparinization was started. The patient remained oliguric despite improvement of systemic hemodynamics. BUN rose from 35 mg% on 7th Dec., 1971 to 68 mg% by 9th Dec., 1971. Plasma creatinine concentration was 1.0 mg% on day of admission, was 2.5 mg% on 8th Dec., 1971, and 4.4 mg% on 9th Dec., 1971. Urine output was 10-20 ml/hr with urine sodium concentrations 35-56 mEq/l. A tentative diagnosis of acute renal failure due to hypotension and sepsis was made.

Selective renal angiography was performed and intrarenal hemodynamics were assessed by the $Xenon^{133}$ washout technique before and after the administration of phenoxybenzamine. Arteriography demonstrated diffusely narrowed intrarenal branches of the renal artery. There was poor perfusion of the renal parenchyma distal to the interlobar level. Following intra-arterial administration of 15 mg of POB over 10 minutes into the right renal artery, there was a considerable increase in the calibre of all renal arteries particularly those in the periphery. There was however, no apparent change in the nephrogram. IRBD by the $Xenon^{133}$ washout technique did not show any change.

The patient remained stable hemodynamically throughout the procedure except for the development of a sinus tachycardia of 100-110/minute. No evidence of improvement of urine output or renal function was evident following intra-arterial phenoxybenzamine blockade.

Seven hours post-angiography, patient suddenly developed respiratory distress, cyanosis and suffered a cardiac arrest. He died 6 days after hospital admission.

Mr. E. G.
WGH 354046

This 74 year old white male who sustained 58% first and second degree burns during a propane gas explosion was transferred from a peripheral hospital to the Winnipeg General Hospital on 21st Oct., 1971. He arrived in Winnipeg after a 6 hour-ambulance journey during which he did not receive any intravenous fluid. He was in clinical shock on arrival. He responded quickly to intravenous albumin-saline resuscitation and was admitted to plastic surgery for the initial care of burns. Initial care was relatively uneventful. The patient was treated with topical gentamycin cream which produced plasma gentamycin levels as high as 4.7 mg/ml at one time. BUN was noted to be elevated by the twelfth hospital day and gentamycin was stopped.

On 5th Nov., 1971 there was increasing sputum and RLL rales and intravenous carbenicillin was started. There was progressive pulmonary and renal functional deterioration and by 15th Nov., 25 days

after hospital admission, the patient developed right sided pneumonia and azotemia. BUN was 100 mg%. The patient was hypotensive, moribund, and required intensive care and endotracheal intubation.

Despite improvement of systemic hemodynamics, stabilization of arterial blood pressure, and improvement of hypoxemia by respiratory support, renal function continued to deteriorate and by 20th Nov., 1971 BUN was 172 mg% with a serum creatinine 9.5 mg%. Urine output remained in the range of 20 ml/hr with a urinary sodium concentration of 63-155 mEq/l.

The cause of acute renal failure developing over a period of 10 days was obscure. The possibility of gentamycin nephrotoxicity was considered. Selective renal angiography was performed on 19th Nov., 1971 and renal vasculature was assessed angiographically as well as by the $Xenon^{133}$ washout method, both before and after the intra-arterial administration of phenoxybenzamine. Extremely poor perfusion of the renal parenchyma was observed both before and after phenoxybenzamine blockade. Phenoxybenzamine did not result in appreciable improvement in cortical perfusion despite a marked increase in the calibre of the more proximal vessels. $Xenon^{133}$ washout studies show similar IRBD before and after POB. Creatinine clearance and urine flow rates showed no significant improvement following blockade.

On 21st Nov., 1971, during the insertion of a central venous

pressure catheter through the internal jugular vein for purposes of hyperalimentation, the patient suffered a cardiac arrest. Resuscitation was unsuccessful. The patient died 30 days after admission to the hospital.

Mr. J. B.
WGH 308048

This 48 year old white male, after the consumption of a considerable quantity of alcohol, fell asleep while smoking in bed on the 29th Oct., 1971. He suffered 60% second and third degree body burns and was brought to the Winnipeg General Hospital 1-2 hours after the accident. On arrival, the patient was hypotensive, with a blood pressure of 80/50 mm Hg, and was in shock. Initial resuscitation by intravenous crystalloid, albumin and fresh frozen plasma was associated with clinical improvement and a rise in urine flow to levels of 200 ml/hr. Because of associated hemoglobinuria, mannitol 25 mg intravenously was given.

The patient was admitted to the plastic surgery service for the initial care of body burns. His respiratory and renal status, deteriorated over the next 5 hours. Urine output decreased to less than 35 ml/hr and became unresponsive to repeated mannitol administration. Urinary sodium concentration measured at this time was 55 mEq/l. A clinical diagnosis of acute renal failure was made. Further volume expansion was not accompanied by a concomittant increase in urine output.

At this time, selective renal angiography was performed which demonstrated extremely poor renal cortical perfusion. Following the intra-arterial administration of 20 mg of phenoxybenzamine over 15 minutes to the right kidney, the more proximal branches of the renal artery showed increases in diameter. Renal cortical perfusion remained poor. Pre- and post-phenoxybenzamine $Xenon^{133}$ washout studies demonstrated findings similar to selective angiography. Intra-arterial phenoxybenzamine was also administered to the left kidney. No improvement of renal function was observed following renal phenoxybenzamine blockade.

General condition of the patient improved with intensive respiratory care. However, 7 hours after transfer to the intensive care unit, the patient aspirated and died of a cardiopulmonary arrest, 20 hours following admission. Resuscitation was unsuccessful. Diagnosis: extensive body burns, pulmonary aspiration and acute renal failure.

Mrs. I. C.
WGH 282623

This 43 year old white female, P viii G viii, was apparently well until May, 1970 when she first noticed swelling of both feet and petechial hemorrhages over lower limbs. Investigation in local hospital raised the possibility of Henoch-Schönlein Purpura. Laboratory findings included: Hb 9.9 gms%; WBC 4,300/mm^3; platelets 326,000/mm^3; reticulocyte count 1.8%; ESR 85 mm/hr; serum Fe 62 mg%; TIBC 522 mg%; BUN 20 mg%; normal urinalysis SGOT 38 units; LAD 80 units. Bone marrow aspirate showed normoblastic hypocellularity of the erythroid series with distortion of chromatin structures.

In July, 1970 patient developed exertional dyspnea, orthopnea and signs of congestive heart failure. Again numerous petechiae over lower extremities were noted. She was readmitted to the local hospital. On 12th Sept., 1970 she developed marked hypotension and dyspnea, hemotypsis and pulmonary edema. She was given digoxin and furosemide. Urinalysis showed trace of albumin, and the presence of granular casts and pus cells. On 18th Sept., 1970 patient showed sudden marked clinical deterioration. She developed generalized abdominal pain, congestive heart failure and oliguria. Laboratory findings were: Hb 12.1 gm%; WBC 16,800/mm^3; ESR 14 mm/hr; BUN 46 mg%. She was transferred to Winnipeg General Hospital on 19th Sept., 1970. By this time, patient

had been anuric for 24 hours. Total urine output was only 5 ml.

On admission, patient complained of marked breathlessness at rest, generalized abdominal pain and vomiting. Physical examination revealed evidence of congestive heart failure and a distended and diffusely tender abdomen with guarding in RLQ. Bowel sounds were absent. Rectal examination was normal. Numerous petechiae were noted in lower extremities, but no edema was present. B. P. was 120/85 mm Hg and pulse was 128/min, regular. BUN was 94 mg%. A diagnosis of congestive heart failure with mesenteric ischemia was considered. Patient continued to be oligo-anuric. Urine sodium concentration at this time was 88 mEq/l.

Superior mesenteric angiogram demonstrated general narrowing and attenuation of the superior mesenteric artery and all of its branches. No vascular occlusion was present. Selective left and right renal angiography was also performed before and after the administration of 5 mg of phenoxybenzamine given by slow infusion over 10 minutes directly into the left renal artery. Before the administration of phenoxybenzamine, the left renal artery was moderately narrowed and the proximal portions of the segmental vessels irregular areas of narrowing, and the nephrogram was exceptionally faint. Post-phenoxybenzamine angiogram showed marked increase in size of virtually all orders of the renal arteries. The nephrogram remained faint and there was no improvement in renal cortical parenchymal perfusion.

In view of the changes in renal vasculature following phenoxybenzamine and the presence of general poor peripheral perfusion, the patient was blocked systemically (2 mg/kg POB). No improvement in renal function was evident, and she continued to deteriorate. Oliguria persisted. On 20th Sept., 1970, BUN was 138 mg%, and plasma creatinine 5.7 mg%. Her abdomen became increasingly tender and rigid. During the insertion of a cordis catheter for peritoneal dialysis, large quantity of fecal-smelling fluid in the peritoneal cavity was noted. Subsequent culture revealed a growth of alpha Streptococcus fecalis 2+, clostridium welchii 2+ and three other clostridium species, none of which were Cl tetani.

Laparotomy was performed on 21st Sept., 1970. Infarction of terminal ileum was found and five feet of the terminal ileum was resected. Her post-operative condition remained poor. The patient was treated with Kanamycin and required the addition of isoproterenol to maintain blood pressure. She developed staphyloccocal septicemia with LLL and RUL infiltrate on 23rd Sept., 1970 and cephalothin was started. Culture from tracheostomy site grew Pseudomonas pyocyanae, resistant to Kanamycin and cephalosporins. Renal function showed no improvement. Peritoneal dialysis was found ineffective and it was changed to hemodialysis. Plasma creatinine varied between 5 and 7 mg%.

On 24th Sept., 1970, patient deteriorated markedly. She developed severe congestive heart failure, pulmonary edema and hypotension. She suffered a cardiac arrest and was successfully resuscitated. On 25th Sept., 1970, she suffered numerous cardiac arrests. Resuscitation was unsuccessful. Patient died on the 6th hospital day.

Post-mortem findings showed myelomatosis, pulmonary arteritis secondary to pseudomonas pneumonia of LLL and RUL, peritonitis. Kidneys appeared somewhat pale and showed mild degenerative tubular changes were present consistent with acute renal failure.

Mr. D.R.
WGH 184326

This 26 year old white male developed childhood nephrosis at the age of 2 years. Progressive deterioration of renal function occurred since that time, finally requiring intermittant hemodialysis in 1968. Bilateral nephrectomies were performed in 1970 for the control of hypertension.

In February, 1971 the patient received a renal cadaveric transplant which was rejected over a period of 2 months. Maintenance hemodialysis was re-instituted.

On the 9th Feb., 1972 a two antigen mismatch cadaveric kidney was transplanted. The operative procedure took approximately 7 hours due to a 360^{o} rotation in the ureter and re-implantation was necessary.

However, the transplanted kidney put out urine within 30 minutes of vascular anastomosis and continued to put out approximately 400 ml/hr of urine for the subsequent 4 hours when urine output suddenly stopped. The patient was returned to the operating room and obstruction of the lower end of the ureter was relieved surgically. Urine output immediately following the second procedure was adequate.

The patient developed oliguria and evidence of rejection over the next five days. This was treated with high doses of steroids. Renal function gradually returned to immediate post-transplant levels over the ensuing 7 days. However, hemodialysis was necessary during the period of oliguria.

On the 28th Feb., 1971, 19 days post-transplant, the patient started to show a decrease in urine output and early signs of rejection. Endogenous creatinine clearance was 49 ml/min. A treatment regime consisting of high doses of steroids and local radiotherapy was started. Renal function progressively deteriorated and on the 5th of March, 1972, 20 days post-transplant, the patient developed marked oliguria. Urine output averaged 10 ml/hr. Endogenous creatinine clearance was 7 ml/min for that day. BUN was 94 mg%. Hemo-

dialysis had to be re-started.

Renal arteriography performed at this time demonstrated an increase in kidney size. All proximal branches of the renal artery appeared normal. However, the arcuate and interlobar vessels appeared to be somewhat stretched. Higher order vessels appeared to be attenuated and showed slight irregularity. No nephrogram could be demonstrated. Intrarenal blood flow distribution as assessed by the $Xenon^{133}$ washout test failed to demonstrate a fast flowing component C_I. Mean renal blood flow was diminished to approximately 1/3 of normal values. Following the intra-arterial administration of 15 mg of Dibenzyline^{R}-SKF (Phenoxybenzamine hydrochloride), angiography and the Xenon washout test was repeated. No significant difference on the angiographic appearance or the Xenon washout curve could be demonstrated.

Oliguria persisted post-phenoxybenzamine administration to as low as 90 ml/day on the 7th of March, 1971. A course of ALG was started and urine output rose to 400 ml/day within 4 days of ALG treatment. Allograft rejection was temporarily under control.

Mrs. D.K.
WGH 211678

This 54 year old white female was admitted to the Winnipeg General Hospital on 14th Nov., 1971 for investigation of paroxysmal explosive vomiting of one week duration. She had a Bilroth II procedure performed in 1965 for peptic disease. A clinical diagnosis of afferent loop syndrome was made.

During her hospital stay, she was noted to be hypertensive with a relatively labile B.P. ranging 200-240/100 - 130 mm Hg. Fundi showed grade ii/iv hypertensive vascular changes. There was no evidence of renal disease. Endogenous C_{Cr} was 83 ml/min. The diagnosis of pheochromocytoma was considered. Biochemical tests including plasma catecholamine concentrations and 24 hour urine for catecholamines were within normal ranges. However, the patient showed an equivocal cold pressor test and a positive glucagon test. Arteriography was performed following systemic phenoxybenzamine (1.5 mg/kg B. Wt.). No tumor was seen in any intra-abdominal site. The diagnosis of pheochromocytoma was temporarily abandoned.

During arteriography the opportunity was utilized for investigation of the renal vasculature. $Xenon^{133}$ washout studies were carried out approximately 16 hours after systemic phenoxybenzamine (1.5 mg/kg) followed by further intra-arterial administration of phenoxybenzamine to the right kidney (10 mg POB given over 10 minutes). The last dose

of POB given intra-arterially was sufficient to block the vasoconstrictor response to an I. A. bolus of adrenaline (16 ug).

Angiographic appearance demonstrated normal vasculature before phenoxybenzamine blockade. Results of $Xenon^{133}$ washout studies are shown in Table XXVIII.

Patient was discharged and was readmitted again in January, 1972 for surgery. The Billroth II was converted to a Billroth I procedure. She was discharged in good condition.