Changes in Renal Blood Flow During Renal Nerve Stimulation[1] (35215)

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The effects of direct renal nerve stimulation (RNS) on renal blood flow (RBF) have been studied by a number of investigators (1–3). However, most studies involved indirect measurements of RBF by clearance techniques which (a) limit measurements to steady state conditions, and (b) require arterial and renal venous blood samples so that repeated measurements in the same animal could result in modest decreases of circulating blood volume. Although excellent direct methods for measuring RBF are available (4), such procedures do not permit facile and accurate examination of RBF changes during commencement, maintenance, and cessation of RNS.

In the present study, RBF changes during direct RNS were measured with an electromagnetic flowmeter. This technique permits examination of transient and sustained flow changes during RNS over a wide range of frequencies in the presence and absence of autonomic blocking drugs. The results show that extent of renal vasoconstriction is directly proportional to frequency of RNS at frequencies between 1 and 10 cycles per second (cps), maximal at 10 cps, and inversely proportional to frequency at frequencies greater than 10 cps. They also show that RBF returns to control flow levels during continued RNS at frequencies above 10 cps. Renal vasoconstrictor responses during RNS appear to be due to activation of alpha-adrenergic receptors, while beta-adrenergic and cholenergic receptors do not seem to be involved. No evidence for extrinsic renal vasodilator innervation was obtained.

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Methods. Experiments were performed in 20 mongrel dogs (15–20 kg), of either sex, anesthetized with sodium pentobarbital (30 mg/kg, iv 30–60 min. following subcutaneous injection of morphine sulfate (3 mg/kg). Each animal breathed spontaneously throughout the experiment. Central arterial and venous pressures were continuously monitored through catheters inserted into the right femoral artery and vein and advanced centrally until their tips were close to the heart. The catheters were attached to Statham P23AC pressure transducers which were connected to a Grass Model 5 polygraph.

The left renal artery was exposed through a retroperitoneal incision. RBF was measured with a noncannulating electromagnetic flow sensor (Statham-Medicon) of the gated sine wave type. Zero flow base line was determined at the beginning of each experiment and checked at the termination of the experiment by occluding the artery distal to the flow sensor.

The renal nerves were isolated and tied with two silk ligatures. The nerve bundle was severed between the ligatures, kept warm and moist, and the peripheral end was stimulated with platinum electrodes. Square wave stimuli of 2-msec duration and variable frequencies (1–200 cps) were delivered from a Grass SD 5 stimulator. In each experiment, voltage (15–30 V) was adjusted until a maximal response occurred at 10 cps. The duration of each stimulus trial (20–30 sec), and elapsed time between trials (1–2 min) was constant in each experiment.

Autonomic blocking drugs, dissolved in saline, were injected (1 ml) into the renal artery through a 25-gauge needle inserted proximal to the flow sensor. The drugs used were phentolamine, propranolol, hexamethonium bromide, and atropine sulfate.

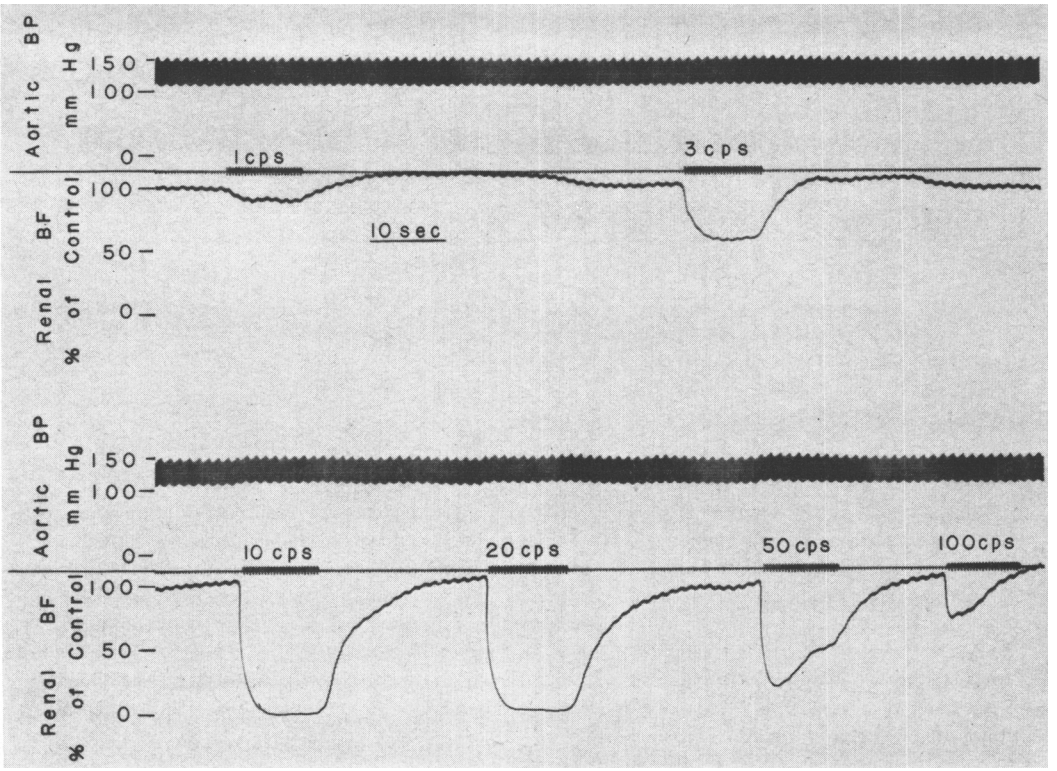

FIG. 1. Effects of renal nerve stimulation on renal blood flow: Supramaximal stimuli of variable frequency (20 V, 2-msec pulse duration) were applied to the renal nerves for the periods indicated on the record. In each panel, the upper trace is arterial blood pressure and the bottom is renal arterial blood flow.

Blood flow changes during RNS were also studied in 3 dogs treated with reserpine (Ciba, 0.5 mg/kg of body weight, subcutaneously) 24 hr prior to study to deplete neural stores of catecholamines (5).

In all experiments, the maximum change in blood flow produced by RNS was expressed as percentage of control RBF measured just prior to stimulation. Statistical evaluation of responses was performed with the Student's t test (6).

Results. The decrease in RBF during RNS was directly proportional to frequency up to 10 cps, but was inversely proportional to higher frequencies of stimulation (Fig. 1). At low frequencies (<10 cps) a transient increase in flow usually occurred following cessation of stimulation. At higher frequencies, such as 50 or 200 cps, RBF often returned to control flow level in the face of continued stimulation. If, during the return toward control flow level, stimulation frequency was reduced, RBF again decreased for the duration of the stimulus trial (Fig. 2). In each experiment decreases in RBF during RNS occurred in the absence of significant changes in arterial blood pressure.

At 1 cps, RBF was reduced to 88 $\pm$ 6% of control (12 dogs, $p<.01$); while at 7.5 cps, RBF decreased to 8 $\pm$ 2% of control ($p<.001$). In all dogs studied, RNS with 10 cps completely shut off the renal blood supply (Table I). Smaller decreases in RBF occurred at higher stimulation frequencies. Thus, at 20 cps, RBF was reduced to 14 $\pm$ 5% of control (17 dogs); while at 100 cps, flow was reduced only to 46 $\pm$ 8% of control (9 dogs, $p<.01$).

Alpha-adrenergic blockade with phentolamine significantly reduced ($p<.001$) RBF responses during nerve stimulation at 5, 10, and 20 cps, while beta-adrenergic blockade

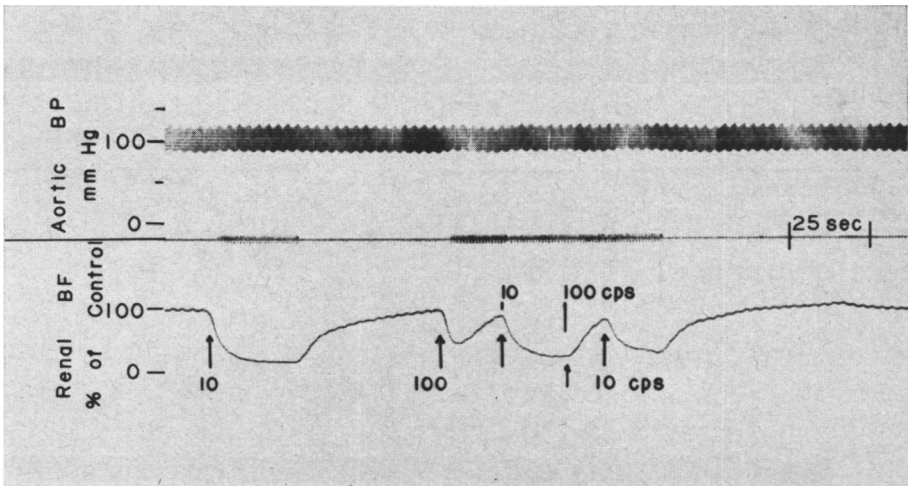

FIG. 2. Renal blood flow response to rapid changes in frequency of renal nerve stimulation: Supramaximal stimuli were applied (20 V, 2-msec pulse duration) for each of the two stimulus trials indicated on the time marker. In the first trial, renal blood flow was decreased to almost zero flow at 10 cps. In the second continuous stimulus trial, stimulation at 100 cps reduced blood flow to only 50% of control but blood flow returned to control as stimulation was continued. Switching frequency to 10 cps caused a prompt decrease in blood flow, but switching back to 100 cps again caused flow to return to control. When frequency was returned to 10 cps, renal blood flow again decreased. High frequency renal nerve stimulation did not appear to deplete neural stores of constrictor substance (norepinephrine).

with propranolol, or ganglionic blockade with hexamethonium did not significantly alter RBF responses (Table I). The dose of propranolol was sufficient to block femoral dilator responses to isoproterenol (2 μg, ia), while hexamethonium blocked the renal vasoconstrictor action of dimethylphenylpiperazinum iodide (DMPP, 50 mg, ia). Atropine significantly attenuated ($p<.01$) RBF decreases during RNS at each frequency tested. Decreases in RBF did not occur when renal nerves were stimulated in reserpinized dogs. In none of the 20 dogs studied was an increase in RBF ever observed during RNS.

Discussion. The frequency–response behavior of the canine renal vasculature (Fig. 1, Table I) during RNS agrees with results reported by Celander working with cats (7). The increase in RBF following cessation of RNS at low frequency shows that the vasculature was not maximally dilated. This poststimulatory hyperemia could be due to metabolic vasodilator substances accumulating during the period of stimulation (8). Return of RBF to control flow levels during continued RNS at high frequenceis (>10 cps) ap-

TABLE I. Effect of Renal Nerve Stimulation on Renal Blood Flow.[a]

Test condition	N[b]	Cycles per second (cps)			
		3	5	10	20
Control	17	67 ± 6	26 ± 5	0 ± 0	14 ± 5
Reserpinized (0.5 mg/kg)	3	98 ± 1	97 ± 2	95 ± 1	99 ± 1
Phentolamine (5 mg, ia)	5		96 ± 3	88 ± 5	80 ± 7
Propranolol (5 mg, ia)	4		21 ± 14	5 ± 4	18 ± 9
Hexamethonium (4 mg, ia)	4		33 ± 5	2 ± 1	9 ± 6
Atropine (2 mg, ia)	4		59 ± 11	26 ± 7	43 ±12

[a] Values are expressed as percentage of control and represent means ± 1 SE.
[b] Number of dogs.

pears to be similar to autoregulatory escape which occurs during nerve stimulation in the intestinal vasculature (9). Renal autoregulatory escape cannot be due to rapid depletion of neural catecholamine stores since switching to lower frequencies of stimulation immediately reduces RBF (Fig. 2). Decreased vasoconstriction observed at high stimulation frequencies (>10 cps) could be related to the finding that release of norepinephrine from sympathetic fibers decreases at high frequency of stimulation (10).

The results shown in Table I are consonant with the following conclusions. First, elimination of renal vasoconstrictor responses during RNS in reserpinized animals is consistent with the concept that catecholamines are required for nerve stimulation to produce vasoconstriction. Second, blockade of constrictor responses with phentolamine supports the view that alpha-adrenergic receptors are activated as a result of RNS. Third, inability to demonstrate increases in RBF during nerve stimulation following alpha-adrenergic blockade, or in reserpinized animals, suggests that extrinsic dilator fibers to the renal vasculature do not exist. Fourth, since propranolol did not alter responses during RNS, beta-adrenergic receptors do not appear to be activated. This finding contrasts with the reported role of beta-adrenergic receptors in the vasculature of the canine gracilis muscle (11), but is consistent with the hypothesis that few responsive beta-adrenergic receptors are present in the renal circulation (12, 13). Fifth, although Page and McCubbin (14) suggested that intrarenal sympathetic ganglia exist, inability of hexamethonium to influence constrictor responses during RNS, indicates that such ganglia, if they exist, do not participate in response to RNS.

The mechanism for attenuation of constrictor responses following atropine administration is unknown. However, similar results were reported by McGiff *et al.* (15).

Summary. Changes in canine renal blood flow (RBF) during direct renal nerve stimulation (RNS) were measured with an electromagnetic flowmeter in the presence and absence of autonomic blocking drugs. The degree of renal vasoconstriction increases as frequency of RNS is increased from 2 to 10 cps. Above 20 cps the degree of vasoconstriction decreases. During continued RNS at high frequencies (>10 cps) RBF returns to control flow level. Renal vasoconstriction during RNS is blocked by alpha-adrenergic blockade, unaltered by beta-adrenergic or ganglionic blockade, and absent in reserpinized animals. These findings suggest that decrease in RBF during RNS is due to activation of alpha-adrenergic receptors by norepinephrine and that the pathway between constrictor fibers and renal vascular smooth muscle probably does not involve ganglionic synapses. No evidence for participation of cholinergic, or beta-adrenergic receptors during RBF responses to RNS, or for the occurrence of extrinsic renal vasodilator nerves was obtained.

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