Furosemide-induced vasodilation: Importance of the state of hydration and filtration

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Furosemide-induced vasodilation: Importance of the state of hydration and filtration. The circumstances under which furosemide increases renal blood flow was examined in mongrel dogs as it may relate to a tubuloglomerular feedback mechanism. Two maneuvers, desoxycorticosterone (DOCA) plus salt treatment and inhibition of tubular fluid flow, were used in the dogs to evaluate the renal vascular effects of furosemide because these maneuvers have been reported to blunt the tubuloglomerular feedback in micropuncture studies. In addition, we also used two structurally different nonsteroidal antiinflammatory drugs to assess the importance of prostaglandins to achieve furosemide's renal vasodilation. Furosemide (5 mg/kg, i.v.) increased renal blood flow in volume-depleted animals from a baseline flow of 141 ± 28 ml/min to a maximum of 176 ± 35 ml/min at 6 min after furosemide administration. If the animals were pretreated with a high-salt diet and i.m. DOCA for 5 days, furosemide administration produced no renal vascular effects but still caused a large diuresis, and these dogs still had a responsive renal vascular bed to infused prostaglandin E_2. In addition, kidneys rendered nonfiltering in volume-depleted animals had no renal vascular response to furosemide. Volume-depleted animals, pretreated with either indomethacin or sodium meclofenamate, did not have a renal vascular response to furosemide although they did have a diuretic response and a responsive renal vasculature to prostaglandin E_2. From our data, we hypothesize that the renal vascular response to furosemide is secondary to a tubular mechanism mediated by a vasodilatory prostaglandin. Because furosemide has been shown to disrupt the tubuloglomerular feedback mechanism, and the two maneuvers, DOCA plus salt treatment and lack of filtration, blunt the tubuloglomerular feedback response as well as inhibit the renal vascular response to furosemide, we further hypothesize that furosemide-induced renal vasodilation may be secondary to the disruption of an active tubuloglomerular feedback mechanism.

Vasodilatation induite par le furosémide: Importance de l'état d'hydratation et de filtration. Les circonstances où le furosémide augmente le débit sanguin rénal ont été étudiées chez des chiens bâtards en fonction d'un mécanisme possible de rétro-contrôle tubulo-glomérulaire. Deux manoeuvres, le traitement par la désoxycorticostérone (DOCA) et le sel et l'inhibition du débit tubulaire, ont été utilisées chez des chiens pour évaluer les effets vasculaires rénaux du furosémide, puisque ces deux manoeuvres sont réputées effacer le rétro-contrôle tubulo-glomérulaire dans les études par microponction. De plus, nous avons employé deux drogues anti-inflammatoires, non stéroïdiennes, de structures différentes pour évaluer l'importance des prostaglandines dans le déterminisme d'une vasodilatation rénale. Le furosémide (5 mg/kg, i.v.) augmente le débit sanguin rénal chez les animaux déshydratés à partir d'une ligne de base de 141 ± 28 ml/min jusqu'à un maximum de 176 ± 35 ml/min 6 minutes après l'administration de furosémide. Quand les animaux ont été prétraités par une alimentation riche en sel et de la DOCA i.m. pendant 5 jours l'administration de furosémide n'a pas produit d'effets vasculaires rénaux mais a cependant déterminé une diurèse importante et le lit vasculaire rénal de ces chiens pouvait encore répondre à la perfusion de prostaglandines E_2. De plus, des reins devenus non filtrants chez des animaux déshydratés n'avaient pas de réponse vasculaire rénale au furosémide. Les animaux déshydratés et pré-traités soit par l'indométhacine, soit par le méclofénamate de sodium, n'avaient pas de réponse vasculaire rénale au furosémide quoiqu'ils avaient une réponse diurétique et une réponse vasculaire rénale à la prostaglandine E_2. De ces résultats nous tirons l'hypothèse que la réponse vasculaire rénale au furosémide est secondaire à un mécanisme tubulaire dont la vasodilatation par la prostaglandine est un médiateur. Puisqu'il a été montré que le furosémide supprime le rétro-contrôle tubulo-glomérulaire et que les deux manoeuvres, DOCA et sel, d'une part, absence de filtration, d'autre part, annulent le rétro-contrôle tubulo-glomérulaire de même qu'elles inhibent la réponse vasculaire rénale au furosémide, nous faisons l'hypothèse supplémentaire que la vasodilatation rénale induite par le furosémide peut être secondaire à l'interruption d'un mécanisme actif de rétro-contrôle tubulo-glomérulaire.

Furosemide, a diuretic derived from anthranilic acid, can increase renal blood flow and enhance renal renin release [1, 2]. The mechanism by which furosemide increases renal blood flow is unclear; two theories can be proposed, however, from the literature. One suggests that furosemide increases renal prostaglandin production, which in turn results in an increased renal blood flow. This theory is supported by the observation that indomethacin can block the renal hemodynamic changes of furosemide [3]; but Duchin, Peterson, and Burke [4] have failed to confirm these findings. The other theory suggests that furosemide acts directly on vascular smooth muscle to produce renal vasodilation [4–6]. It seems unlikely, however, that furosemide direct-

ly affects vascular smooth muscle because not all vascular beds respond qualitatively the same way as the kidney [7, 8].

Another possible mechanism whereby furosemide could affect renal vascular resistance is through interruption of the tubuloglomerular feedback mechanism. Recently, Briggs and Wright have reported that the effector mechanism for the feedback control of the GFR in a single nephron is afferent arteriole constriction [9]. Thus, interruption of this pathway in all nephrons would result in afferent arteriolar dilation with a resultant increase in renal blood flow. Indeed, Wright and Schnermann have reported that furosemide interferes with the tubuloglomerular feedback mechanism in micropuncture experiments [10]. In addition, we have evidence that the renal vasoconstriction produced by an infusion of hypertonic saline into the renal artery is somewhat analogous to the tubuloglomerular mechanism in isolated nephrons inasmuch as both are inhibited by the administration of desoxycorticosterone (DOCA), furosemide, and lack of filtration [11].

The current experiments were designed in dogs to study the effect of hydration plus DOCA administration and the lack of tubular fluid flow on the renal vascular effects of furosemide. These experimental circumstances were chosen because they have been reported to blunt the tubuloglomerular feedback mechanism in micropuncture experiments [12]. In addition, we further explored the importance of vasodilatory prostaglandins in mediating the decrease in vascular resistance produced by furosemide.

Methods

Twenty-two mongrel dogs of either sex weighing 18 to 30 kg were used to assess the renal hemodynamic effects of furosemide (5 mg/kg, i.v.). The dogs were divided into four experimental groups.

Group 1. Six mongrel dogs were acutely volume-depleted by depriving them of food and water for 24 hours prior to the experiment. On the day of the experiments, the dogs were anesthetized with pentobarbital (30 mg/kg, i.v.) and respired using a positive pressure respiratory pump at a rate of 16 breaths/min. A femoral artery was catheterized for blood pressure monitoring, and a femoral vein was catheterized for drug administration.

Through a midline abdominal incision, the left renal artery was identified and cannulated with a 23-guage needle for the infusion of hypertonic saline. A noncannulating electromagnetic flow probe was placed around the renal artery distal to the needle. The left ureter was catheterized for urine collection. After the animal had stabilized, hypertonic sodium chloride, calculated to raise the renal arterial plasma concentration of sodium by 30 mEq/liter, was administered into the left renal artery for a total of 10 min, and the renal vasoconstrictor response was determined. We have reported the analogy between this vasoconstrictor response in the whole kidney, and the tubuloglomerular feedback mechanism in micropuncture experiments because both were inhibited by hydration plus DOCA, furosemide, and lack of tubular fluid flow [11]. When the renal blood flow, urine flow, and blood pressure had returned to baseline (usually 45 min), furosemide (5 mg/kg, i.v.) was administered, and renal blood flow and arterial blood pressure were monitored continuously. Urine was collected for the first 15 min after furosemide and subsequently analyzed for sodium and potassium by flame photometry.

Group 2. Five mongrel dogs were placed on diets containing 250 mEq of sodium per day combined with daily injections of 10 mg of desoxycorticosterone (DOCA) for a total of 5 days prior to the experiments. On the day of the experiment, the dogs were fasted overnight, and given their DOCA injection early in the morning. The dogs were then surgically prepared identically to group 1 dogs. They too underwent hypertonic saline infusion into the renal artery as described with group 1 followed by i.v. furosemide. The renal blood flow, arterial blood pressure, and urinary output of sodium and potassium were monitored before and after furosemide as described before.

Group 3. Six mongrel dogs were dehydrated and surgically prepared as described for dogs in group 1 except that these dogs were randomly assigned to pretreatment with indomethacin (8 mg/kg, i.v.) or sodium meclofenamate (10 mg/kg, i.v.) 30 min prior to the infusion of the hypertonic saline (three dogs received indomethacin, and three dogs received meclofenamate). The rest of the protocol was followed as with the group 1 dogs with renal blood flow, arterial blood pressure, and urinary output of sodium and potassium monitored before and after furosemide.

Group 4. Five mongrel dogs had one kidney rendered nonfiltering by the procedure described by Blaine, Davis, and Witty [13]. The animals were also acutely volume depleted as described in the previous sections. On the day of the experiment, the animals were anesthetized with sodium pentobarbital (30 mg/kg, i.v.) and prepared surgically as de-
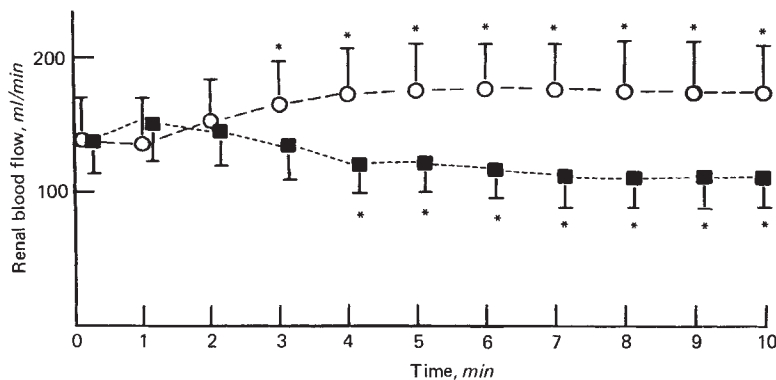
Fig. 1. *Effect of furosemide and intrarenal hypertonic saline on the renal blood flow in dehydrated animals.* The *open circle* represents the renal blood flow after furosemide administration, and the *closed boxes* represent renal blood flow after intrarenal hypertonic saline infusion. The *asterisk* signifies that the change in renal blood flow is significantly different ($P < 0.05$) from the control blood flow. The *zero value* represents the control blood flow. Values represent the means ± SEM ($N = 6$).

scribed for dogs in group 1. An electromagnetic flow probe was placed around the renal artery of the nonfiltering kidney, and renal blood flow was continuously monitored. The filtering kidney was surgically removed. These dogs were also given furosemide (5 mg/kg, i.v.). After the termination of the experiments, 2 ml of indigo carmine was infused i.v. to confirm that the kidney was nonfiltering.

Statistics. The renal hemodynamic data were analyzed by Dunnett's t test, comparing control blood flow with the multiple blood flow measurements after furosemide administration. A P value of less than 0.05 was considered statistically significant.

Results

Group 1. Furosemide, in the volume-depleted animals, significantly increased renal blood flow starting at 3 min after the drug administration and reaching a maximum at 6 min (Fig. 1). In these animals, there was a significant decrease in renal blood flow during the intrarenal infusion of hypertonic saline infusion. Associated with furosemide's vasodilation, there was a marked diuresis, natriuresis, and kaliuresis (Table 1).

Group 2. In the salt-loaded and DOCA-treated animals, furosemide had no renal vasodilating effect. There was a modest decrease in renal blood flow at 1 min but for the remaining 9 min the renal blood flow was not any different from baseline (Fig. 2). To document that the renal vasculature could still respond to a vasodilatory stimulus, we infused prostaglandins E_2 (PGE_2) at a dose of 100 ng/kg/min into the renal artery, which resulted in a significant increase in renal blood flow. In all these animals, unlike in the volume-depleted animals, hypertonic saline infusion into the renal artery produced only increases in renal blood flow. Intravenous furosemide in all these animals also resulted in a marked natriuresis (Table 1).

Group 3. Indomethacin and sodium meclofenamate did not interfere with the renal vasoconstriction produced by an intrarenal infusion of hypertonic saline in these volume-depleted animals; these prostaglandin synthesis inhibitors, however, completely blocked the renal vascular effects of furosemide (Fig. 3). The kidneys still had a responsive vasculature after the administration of the nonsteroidal antiinflammatory drugs, as seen by the

**Table 1.** The effect of furosemide (F) on urinary sodium and potassium excretion[a]

	Volume, *ml/min*		Sodium, *μEq/min*		Potassium, *μEq/min*	
	Before F	After F	Before F	After F	Before F	After F
Dehydrated dogs ($N = 6$)	0.11 ± 0.04	3.7 ± 0.58[c]	17 ± 5	468 ± 53[d]	21 ± 7	104 ± 13[c]
DOCA + salt-treated dogs ($N = 5$)	0.73 ± 0.22	5.7 ± 1.7[b]	156 ± 68	936 ± 268[b]	25 ± 5	102 ± 20[b]
NSAI (meclo- or indo-treated dehydrated dogs)	0.05 ± 0.01	1.1 ± 0.3[b]	8 ± 3	196 ± 35[c]	11 ± 4	74 ± 18[c]

[a] Values are the means ± SEM. Abbreviations used are DOCA, desoxycorticosterone; NSAI, nonsteroidal antiinflammatory drugs; meclo, sodium meclofenamate; indo, indomethacin.
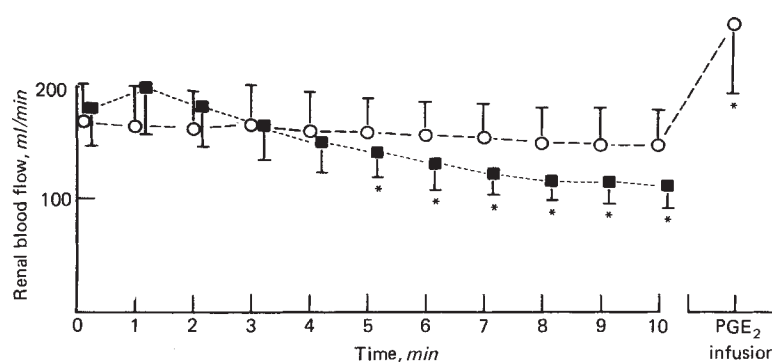[b] $P < 0.05$.
[c] $P < 0.01$.
[d] $P < 0.001$.
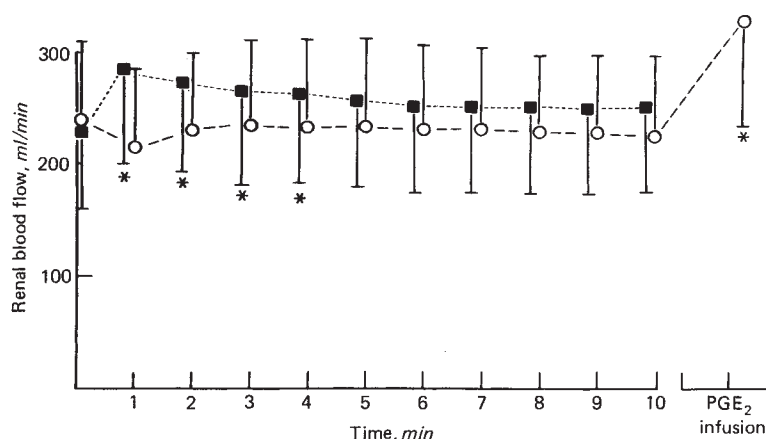
Fig. 2. *Effect of furosemide and intrarenal hypertonic saline on the renal blood flow in salt-loaded and DOCA-treated dogs.* The *open circle* represents the renal blood flow after furosemide administration, and the *closed box* represents renal blood flow after intrarenal hypertonic saline infusion. The *asterisk* signifies that the change in renal blood flow is significantly different ($P < 0.05$) from the control blood flow. Prostaglandin E_2 was administered intrarenally to assess whether the renal vascular bed was responsive to vasodilatory prostaglandin stimulus. Values represent means ± SEM ($N = 5$).

Fig. 3. *Effect of furosemide and intrarenal hypertonic saline on the renal blood flow in prostaglandin-inhibited animals.* The *open circle* represents renal blood flow after furosemide administration, and the *closed box* represents renal blood flow after intrarenal hypertonic saline infusion. The *asterisk* signifies that the change in renal blood flow is significantly different ($P < 0.05$) from the control blood flow. Prostaglandin E_2 was administered intrarenally to assess whether the renal vascular bed could respond to a vasodilatory prostaglandin stimulus. Values represent means ± SEM ($N = 6$).

large increase in renal blood flow produced by intrarenal PGE_2 infusion (100 ng/kg/min). The nonsteroidal antiinflammatory drugs altered the diuretic effect of furosemide very little (Table 1).

Furosemide did not produce renal vascular changes in the nonfiltering kidneys of volume-depleted dogs (Fig. 4). In none of the animals did furosemide produce consistent effects on arterial pressure (Table 2).

Discussion

Although the renal vasoactivity of furosemide was described over a decade ago, the exact mechanism of this vasodilation is still unknown. It is clear that not all investigators have found that furosemide increases renal blood flow, and not all vascular beds

respond to furosemide in the same manner as the renal vasculature [7, 8, 14, 15]. These findings probably indicate that the renal vasodilation is not a direct effect of furosemide on vascular smooth muscle and that the vascular response occurs only under certain circumstances. Our present data show that the renal vascular effect of furosemide requires prostaglandin synthesis and renal tubular fluid flow, and that the vascular response to furosemide is inhibited by pretreatment with DOCA and salt, a maneuver that does not block the renal vasodilator response to PGE_2.

Data from micropuncture experiments indicate that the renal tubule can control renal afferent arteriolar tone through a feedback mechanism, and furosemide can interrupt this mechanism [9, 10]. We

Table 2. Blood pressure effects of furosemide (in mm Hg)[a]

	Dehydrated dogs (N = 6)	DOCA + salt (N = 5)	NSAI-treated, dehydrated (N = 6)	Nonfiltering, dehydrated (N = 5)
Before F	135 ± 4	136 ± 3	120 ± 7	123 ± 4
After F	130 ± 3	135 ± 3	121 ± 7	123 ± 3

[a] Values are the means ± SEM. Abbreviations are defined in Table 1. None of the changes was statistically significant.

postulate that the furosemide increases renal blood flow in the whole kidney by a similar mechanism, that is, by interruption of a tubuloglomerular feedback. Our data support this hypothesis because the two maneuvers, volume expansion plus DOCA and the lack of renal tubular fluid movement, not only inhibited furosemide's renal vascular effect in our experiments but also are known to blunt the tubuloglomerular feedback mechanism. In addition, all the diuretics that act like furosemide to inhibit ion transport in the loop of Henle have been reported to produce renal vasodilation. Thiazide diuretics that inhibit ion transport beyond the loop of Henle do not decrease renal vascular resistance [16].

Our proposal that furosemide vasodilates the kidney through the inhibition of the tubuloglomerular feedback is purely deductive because we have no micropuncture evidence that we are, indeed, inhibiting the tubuloglomerular feedback mechanism by furosemide. The parallel conditions of inhibition in our study and those described in the micropuncture literature suggest, however, that furosemide's vascular effects are through a tubular rather than a direct vascular mechanism. It is of interest that an infusion of hypertonic saline into the renal artery produced renal vasoconstriction only in those animals that also had a renal vasodilator response to furosemide. The renal vascular response to hypertonic sa-

line also shows similarities to the activation of a tubuloglomerular feedback in that it is inhibited by DOCA-salt treatment and by furosemide [11]. Although most of the literature dealing with the tubuloglomerular feedback mechanism is in rats, Navar et al have shown in micropuncture studies that dogs also have an active tubuloglomerular feedback mechanism [17].

The mediator for the renal vasodilation following furosemide administration is a prostaglandin because two structurally dissimilar inhibitors of prostaglandin synthesis were equally proficient in inhibiting this vascular response. These findings are in agreement with Bailie, Barbour, and Hook [3] and Data et al [18] but are in disagreement with Duchin, Peterson, and Burke [4]. The discrepancy between our findings and the findings of Duchin et al is not readily explicable, but it is clear from our data that the nonsteroidal antiinflammatory drugs did not alter renal vascular responsiveness because the kidney still vasodilated to PGE_2. The data also agree with Bailie, Crosslan, and Hook [19] that prostaglandin inhibition does not alter the diuretic response to furosemide; therefore furosemide's diuretic effect is neither prostaglandin nor hemodynamically mediated.

If our hypothesis that furosemide produces renal vasodilation by interrupting an active tubuloglomerular feedback is corroborated by micropuncture experiments, we would predict that all drugs and interventions that inhibit the tubuloglomerular feedback mechanism should inhibit furosemide's vascular effect as well. Such studies should help to reconcile the reported discrepancies in the literature regarding the vascular effects of furosemide.
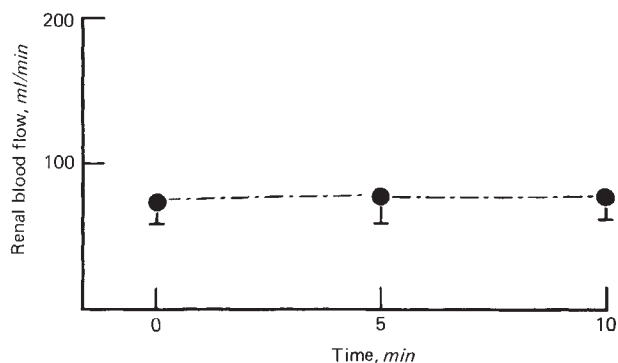
Fig. 4. *Effect of furosemide on renal blood flow in animals with a single nonfiltering kidney (●, N = 5). Values represent means ±* SEM.

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