Urocortin 2 Inhibits Furosemide-Induced Activation of Renin and Enhances Renal Function and Diuretic Responsiveness in Experimental Heart Failure

Miriam T. Rademaker, PhD; Christopher J. Charles, PhD; M. Gary Nicholls, MD, FRACP; A. Mark Richards, MD, PhD, FRACP

Background—Urocortin 2 (Ucn2), a novel peptide with therapeutic potential in heart failure, and diuretics have opposing effects on renal function and the renin-angiotensin-aldosterone system. Because any prospective new treatment is likely to be used in conjunction with standard diuretic therapy, it is necessary to investigate the combined effects of these agents.

Methods and Results—Ucn2 and furosemide were administered for 3 hours, both singly and combined, in 7 sheep with pacing-induced heart failure. Compared with time-matched controls, separate Ucn2 and furosemide administration significantly increased urine output (furosemide>Ucn2), urine sodium (furosemide>Ucn2), potassium (furosemide>Ucn2), and creatinine excretion (Ucn2>furosemide) and creatinine clearance (Ucn2>furosemide). Compared with furosemide treatment alone, Ucn2+furosemide produced a further diuresis (P<0.05), natriuresis (P<0.05), and a sustained increase in creatinine excretion (P<0.05) and clearance (P<0.05), without additional potassium elimination. All active treatments reduced mean arterial pressure (Ucn2+furosemide=furosemide>Ucn2), left atrial pressure (Ucn2+furosemide>Ucn2>furosemide), and peripheral resistance (Ucn2+furosemide=Ucn2>furosemide), whereas only Ucn2, singly and in combination with furosemide, increased cardiac output and dP/dt(max). In contrast to the increase in plasma renin activity elicited by furosemide alone, Ucn2 and Ucn2+furosemide markedly reduced plasma renin activity. All active treatments decreased plasma aldosterone (Ucn2+furosemide=Ucn2>furosemide), whereas only Ucn2 and Ucn2+furosemide reduced vasopressin and natriuretic peptide concentrations.

Conclusion—Ucn2 cotreatment with furosemide enhanced hemodynamic and renal function and diuretic responsiveness (without additional potassium depletion) in experimental heart failure. Furthermore, Ucn2 reversed furosemide-induced increases in plasma renin activity and induced greater decreases in plasma aldosterone and vasopressin. These data indicate that adjunct Ucn2 therapy with diuretics in heart failure is beneficial. (Circ Heart Fail. 2009;2:532-540.)

Key Words: urocortin 2 ▪ diuretics ▪ heart failure ▪ hemodynamics ▪ hormones ▪ renal function

Urocortin 2 (Ucn2) is a novel vasoactive peptide belonging to the corticotropin-releasing factor family that acts through the receptor subtype corticotropin-releasing factor type 2.1 Both Ucn2 and its receptor are found in high concentrations in the heart and blood vessels,1,2 and the cardiovascular effects of the peptide in normal animals and man, including vasodilator, lusitropic, and positive cardiac contractile actions,2–5 suggest that Ucn2 participates in the regulation of pressure homeostasis in health. In experimental and human heart failure, Ucn2 enhances left ventricular ejection fraction and cardiac output (CO) in association with reductions in systemic peripheral resistance, blood pressure, left atrial pressure (LAP), and cardiac work.3,4,6 In addition to its hemodynamic effects, the peptide is reported to inhibit several vasoconstrictor/volume-retaining hormone systems (including the renin-angiotensin system [RAS] and aldosterone secretion) and improve renal function (increasing urine output, sodium and creatinine excretion, and creatinine clearance) in an ovine model of congestive heart failure (CHF).4 In patients with CHF, creatinine excretion was well maintained in the face of marked Ucn2-induced decreases in arterial pressure (=20 mm Hg) in association with relative suppression of the RAS.8 Furthermore, the peptide has been shown to reduce both infarct size and arrhythmias after ischemia/reperfusion injury in rats.7 These findings suggest that Ucn2 may play a role in the pathophysiology of CHF and reveal a therapeutic potential for the peptide in this increasingly common disease.

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Currently, a mainstay of therapy for CHF is the use of diuretic agents (especially potent loop diuretics such as furosemide) that primarily target the kidney to promote sodium and water excretion, thereby improving central he-

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modynamics and cardiac pumping secondary to reduced plasma and extracellular volumes. However, although diuretic therapy is valuable in treating congestion, it may in fact worsen renal function. A number of studies have demonstrated that (even low-dose) loop diuretics can reduce glomerular filtration rate, which is reported to be a key predictor of mortality in CHF, as well as lead to deleterious stimulation of the RAS and aldosterone. In addition, over time and in severe CHF, patients often become refractory to diuretics, in which case higher doses (which are associated with a poor prognosis) must be used and/or adjuvant/alternate strategies implemented to reduce salt and water overloading. In fact, the long-term use of loop diuretics in the treatment of CHF has been a matter of considerable debate.

Because any prospective new treatment, such as Ucn2, is likely to be used conjointly with standard diuretic therapy, it is essential that the integrated effects of these agents be reviewed. This is of particular interest in this setting given that the opposing actions of Ucn2 and diuretics on renal function and the RAS and aldosterone, leading us to hypothesize that Ucn2, when coinfused with the loop diuretic furosemide, might enhance the therapeutic efficacy of the latter while inhibiting its undesirable actions. In this study, we examine for the first time the hemodynamic, endocrine, and renal effects of Ucn2 and furosemide, administered both singly and combined, in sheep with experimental CHF.

Methods

Surgical Preparation of Sheep

Seven Coopworth ewes (50 to 60 kg) were instrumented through a left lateral thoracotomy under general anesthesia (induced by 17 mg/kg thiopentone; maintained with halothane/nitrous oxide) as previously described. In brief, 2 polyvinyl chloride catheters were inserted into the left atrium for blood sampling and LAP determination; a Konigsberg pressure-tip transducer was inserted into the aorta to record mean arterial pressure (MAP) and into the apex of the left ventricle to obtain maximum derivatives of pressure over time (dP/dt [max]) as an index of contractility; an electromagnetic flow probe was placed around the ascending aorta to measure CO; a Swan-Ganz catheter was inserted into the pulmonary artery for infusions; and a 7-French His-bundle electrode was stitched subepicardially to the wall of the left ventricle for pacing. A bladder catheter was inserted per urethra to facilitate accurate urine collections. Animals recovered for at least 14 days before starting the study protocol. During the experiments, the animals were held in metabolic cages and had free access to water and food (containing 80 mmol/d sodium; 200 mmol/d potassium).

Study Protocol

CHF was induced by 7 days of rapid left ventricular pacing at 225 bpm and maintained by continuous pacing for the duration of the study. On days 8, 10, 12, and 14 of pacing, each sheep received, in random order, a vehicle control (0.9% saline), murine Ucn2 (50 μg bolus +50 μg/h infusion; American Peptide Company Inc., Sunnyvale, Calif), the loop diuretic furosemide (20 mg bolus+3.3 mg/h infusion; Mayne Pharma Pty Ltd., Mulgrave, Victoria, Australia), and the combination of both agents. All treatments were administered for 3 hours in a total volume of 50 mL through the pulmonary artery catheter. The doses of both Ucn2 and furosemide chosen were based on our earlier experience with these agents in the same animal model.

MAP, LAP, CO, dP/dt(max), and calculated total peripheral resistance (CTPR = MAP/CO) were recorded at 15-minute intervals in the hour before infusion (baseline) and at 15, 30, 45, 60, 90, 120, 150, and 180 minutes during both the 3-hour infusion and postinfusion periods. Hemodynamic measurements were determined by online computer-assisted analysis (PowerLab Systems, ADInstruments, Dunedin, New Zealand).

Blood samples were drawn from the left atrium 30 minutes and immediately preinfusion (baseline) and at 30, 60, 120, and 180 minutes during the 3-hour infusion and postinfusion periods. Samples were taken into EDTA tubes on ice, centrifuged at 4°C, and stored at either −20°C or −80°C before assay for plasma renin activity (PRA), aldosterone, arginine vasopressin (AVP), endothelin-1, atrial natriuretic peptide, brain natriuretic peptide, and catecholamines. For each hormone, all samples from individual animals were measured in the same assay to avoid interassay variability. Plasma electrolytes and hematocrit were measured with every blood sample taken.

Urine was collected hourly for the measurement of volume and sodium, potassium, and creatinine excretion. Creatinine clearance was calculated as urine creatinine/plasma creatinine.

The study protocol was approved by the local University of Otago Animal Ethics Committee.

Statistics

Results are expressed as mean±SEM. Baseline hemodynamic and hormone values represent the mean of the 4 and 2 measurements, respectively, made within the hour immediately pretreatment. Comparison of baseline data by ANOVA showed no significant differences for any variable between the 4 study days or an effect of treatment order, confirming the adequacy of the washout period between treatments and indicating that the animals are stable and returned to baseline in the time interval between study days. Differences between the 4 study days (both treatment and post-treatment phases included) were determined using 2-way repeated-measures ANOVA. Significance was assumed at P<0.05. Treatment×time interactions are quoted in the text unless otherwise stated. Where significant differences were identified by ANOVA, the level of significance at individual time points in Figure 1 and Tables 1 and 2 was determined by Dunnett’s tests.

Results

Rapid left ventricular pacing at 225 bpm for 7 days produced the hemodynamic, endocrine, and sodium-retaining hallmarks of CHF, with reduced CO, MAP and renal function, increased LAP and peripheral resistance, and widespread hormonal activation.

Compared with time-matched control data, all 3 treatments significantly increased urine output (Ucn2, P<0.05; furosemide, P<0.001; Ucn2+furosemide, P<0.001), urine sodium (Ucn2, P<0.05; furosemide, P<0.001; Ucn2+furosemide, P<0.001) and potassium excretion (all P<0.001; Figure 1). However, although furosemide-induced responses (alone or in conjunction with Ucn2) tended to be more immediate and considerably greater, Ucn2-inclusive responses were more sustained. Despite the enhanced drinking in response to furosemide alone (P<0.01; Table 1), administration of this agent (P<0.05), as well as Ucn2 (P<0.05) and Ucn2+furosemide (P<0.001), resulted in significant negative fluid balances (drinking−urine volume) relative to control (Table 1).

Creatinine excretion (Figure 1) and creatinine clearance (Table 1) were markedly increased by Ucn2 for the duration of the 3-hour infusion period and the 1 to 2 hours beyond (Ucn2 both P<0.01; Ucn2+furosemide both P<0.001) and transiently (during the first hour only) by furosemide alone (both P<0.001). All treatments reduced plasma potassium concentrations (Ucn2, P<0.01; furosemide, P<0.001;
Ucn2+furosemide $P<0.001$), whereas Ucn2 alone decreased plasma creatinine ($P<0.05$), and furosemide alone tended to increase plasma creatinine (0.1 $P>0.05$). Compared with the consistent gradual decrease in hematocrit seen over the control day, levels were reduced further during Ucn2 ($P<0.001$) and, to a lesser extent, furosemide administration ($P<0.001$), whereas an intermediate response was produced by Ucn2+furosemide (Table 1). Plasma sodium concentrations were not significantly altered by any treatment (Table 1).

Figure 1. Mean±SEM renal responses to 3-hour infusions of vehicle (open bars), Ucn2 (50 $\mu$g bolus+50 $\mu$g/h infusion; striped bars), furosemide (20 mg bolus+3.3 mg/h infusion; dotted bars), and Ucn2+furosemide combined (closed bars) in 17 sheep with heart failure. Significant differences from time-matched control data are shown by: *$P<0.05$, **$P<0.01$, #$P<0.001$.

Compared with furosemide treatment alone, the combination of Ucn2+furosemide produced a further increase in sodium excretion ($P<0.05$) and urine output ($P<0.05$) (with an associated greater decline in fluid balance [$P<0.05$]) and a sustained increase in creatinine excretion ($P<0.05$) and creatinine clearance ($P<0.05$; Figure 1; Table 1). Ucn2 cotreatment also prevented the increase in plasma creatinine concentrations ($P=0.05$ [treatment effect]). Importantly, potassium excretion was not additionally enhanced beyond that produced by furosemide alone (Figure 1). This is also apparent when looking at total potassium excreted during the 3-hour Ucn2+furosemide infusion period (61.7±10.1 mmol), which, although significantly greater than that excreted during the control (18.9±5.1 mmol, $P<0.05$) and Ucn2 (44.0±6.0 mmol, $P<0.05$) treatments, was equivalent to the furosemide total (60.7±6.9 mmol). On the other hand, total sodium excretion was clearly increased during Ucn2+furosemide administration (242.1±36.85 mmol) relative to all other treatments (control 0.74±0.46, $P<0.001$; Ucn2 27.87±15.99, $P<0.01$; furosemide 168.93±15.68 mmol, $P<0.05$), and total fluid balance was clearly more negative during the combined treatment (~1562±200 mL) compared with the individual agents (control +476±238, $P<0.001$; Ucn2 −150±209, $P<0.01$; furosemide −670±146 mL, $P<0.05$).

Separate Ucn2 and furosemide administration reduced MAP (furosemide>Ucn2; both $P<0.001$), LAP (Ucn2>furosemide; both $P<0.001$), and CTPR (Ucn2>furosemide; both $P<0.001$) (Figure 2). Only Ucn2 induced a significant increase in CO ($P<0.001$; Figure 2) and dP/dt(max) ($P<0.001$; Table 2).

Dual Ucn2+furosemide treatment had similar effects on CTPR ($P<0.001$), CO ($P<0.001$), and dP/dt(max) ($P<0.001$) (Figure 2; Table 2) to those induced by Ucn2 alone, with added reductions in MAP ($P<0.001$) and LAP ($P<0.001$).

In contrast to the increase in PRA observed during furosemide alone ($P<0.05$), Ucn2 and Ucn2+furosemide administration significantly reduced PRA compared with control ($P<0.05$ and $P<0.001$, respectively; Figure 3). All 3 treatments decreased plasma aldosterone levels (Ucn2 $P<0.001$; furosemide $P<0.01$; Ucn2+furosemide $P<0.01$), the declines being greater and more sustained during Ucn2+furosemide than with furosemide alone ($P<0.05$; Figure 3). Ucn2, alone and in combination with furosemide, also reduced plasma AVP (both $P<0.05$ [treatment effects]).
atrial natriuretic peptide (both \(P<0.001\)), and brain natriuretic peptide (both \(P<0.001\)) concentrations (Figure 3; Table 2). Plasma endothelin-1 and catecholamines were not significantly affected by any treatment (Table 2).

**Discussion**

This study is the first to report the effects of combined Ucn2 and diuretic therapy in CHF. We found that, compared with furosemide treatment alone, concomitant administration of Ucn2 produced a further natriuresis and diuresis and increased creatinine clearance. Whereas furosemide alone had little or no effect on CO, LAP, and CTPR, the addition of Ucn2 had sizable beneficial actions on each of these hemodynamic indices. In addition, Ucn2 cotreatment reversed furosemide-induced increases in PRA and induced further reductions in plasma aldosterone and AVP.
Table 2. Effects of Urocortin 2 and Furosemide, Separately and Combined, in 7 Sheep With Heart Failure

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<th></th>
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Values are mean±SEM. Significant differences between time-matched vehicle control and active treatments are shown by: †P<0.01, ‡P<0.001. ANP indicates atrial natriuretic peptide.

An increasing emphasis is now being placed on the role of the renal system in CHF in light of growing evidence that worsening renal function, even a mild deterioration, seems to be prognostic of a poor outcome.10,21 In this study, although the trend for plasma creatinine to increase during short-term furosemide administration was not associated with a (perceptible) decline in creatinine clearance (and presumably glomerular filtration rate), the connection between chronic diuretic therapy and creatinine clearance/glomerular filtration rate diminution is well established, especially in patients with already compromised renal function.9 Cotreatment with Ucn2 in our study abolished the increase in plasma creatinine seen with furosemide alone along with significant and sustained increases in creatinine clearance. In addition, Ucn2 enhanced the diuretic (and natriuretic) responsiveness to furosemide—an important finding given that the development of diuretic resistance (defined clinically as a high dose requirement), which is common among patients with severe CHF, is associated with increased mortality.12,13 Remarkably, these enhanced renal actions with Ucn2 confusion occurred in the face of pronounced reductions (≈70%) in plasma levels of the natriuretic peptides—reductions that were absent during separate furosemide infusion. The positive impact of Ucn2 on glomerular filtration rate (creatinine clearance) and urine volume and sodium excretion has been noted previously in our ovine model of CHF.4 Although the underlying mechanisms are unknown, possible contributing factors include an improvement in CO, renal vasodilatation,22 and perhaps, direct tubular actions in light of Ucn2 gene expression within the kidney.1 In addition, Ucn2-induced attenuation of antinatriuretic/antidiuretic factors, such as angiotensin II (as demonstrated previously23 and judged by increases in PRA in this study), aldosterone, and AVP, may have played a part. It is also possible that Ucn2 cotreatment induced a greater urinary delivery of furosemide (especially in light of the concomitant increase in creatinine clearance) and therein a greater response.

Importantly, the combination of Ucn2 and furosemide did not increase potassium excretion over and above that observed with furosemide separately, a finding reinforced by the similar plasma potassium levels evident with the 2 treatment regimens. This is of clinical relevance given the potential risk...
of hypokalemia with loop diuretics (and the associated proarhythmic effects). Our results indicate that the effects of Ucn2 cotreatment to enhance renal function and diuretic/natriuretic responsiveness (without additional potassium depletion) during furosemide administration in CHF would be beneficial, perhaps most especially in otherwise diuretic-resistant patients with volume overload.

Another major finding in this acute study is the impact of Ucn2 cotreatment on the RAS and aldosterone. Although furosemide increased PRA, a well-established consequence of loop diuretics, Ucn2 coadministration not only prevented the increase in renin activity (and presumably angiotensin II) but also produced impressive and persistent reductions (~70%) in its circulating concentrations. With regard to aldosterone, furosemide alone temporally reduced plasma levels of the steroid in this acute setting—a somewhat unexpected result considering the coincidental increase in PRA. This aldosterone decline is likely attributable to the concomitant (diuretic-induced) decrease in plasma potassium concentrations, given the close relationship evident between changes in circulating potassium (including a sizable 27% reduction) and aldosterone during furosemide administration (with alterations in potassium preceding those of aldosterone). Indeed, the 2 variables are strongly correlated in these diuretic-treated animals (r=0.879, P<0.001). It is also possible that the trend for CO to increase during furosemide administration (P=0.086) may have resulted in improved hepatic clearance of the steroid. Adjunct Ucn2 treatment induced a further decrease in plasma aldosterone that was maintained well beyond the treatment period. These are also likely to be meaningful outcomes clinically because the maladaptive stimulation of the RAS and aldosterone (associated especially with long-term or aggressive diuretic therapy) has deleterious effects to increase systemic vascular resistance, limit sodium excretion (through actions of angiotensin II at the proximal tubule and aldosterone at the distal tubule), and promote cardiovascular remodeling long term, leading to accelerated morbidity and mortality. Not surprisingly, given their renal and vascular effects, RAS and aldosterone activation is reported to be a major risk factor in diuretic resistance. Whether attenuation of circulating PRA levels by Ucn2, also observed in our earlier studies in experimental CHF, is due to direct inhibition of renin secretion/release, increased sodium (and chloride) delivery to the macula densa, a decline in sympathetic drive to the juxtaglomerular cells, or some other renin-suppressive mechanism is unknown. On the other hand, Ucn2-induced reductions in plasma aldosterone levels in this study likely reflect decreased plasma angioten-
sin II$^{23}$ (as judged by the decrease in PRA) and potassium levels, although a direct inhibitory effect on aldosterone secretion is also a possibility given the presence of Ucn2 message expression in the adrenal gland.$^{1}$ The effect of Ucn2 on renal tubular transport processes, which can also play a role in diuretic resistance,$^{27}$ is yet to be investigated.

A significant difference was demonstrated in the plasma natriuretic peptide response between the furosemide and Ucn2$^{3}$ groups. Whereas furosemide treatment did not alter circulating atrial natriuretic peptide and brain natriuretic peptide levels, the addition of Ucn2 produced pronounced decreases in plasma levels of the peptides (equivalent to those seen during Ucn2 administration alone). We have observed similar effects during previous work with Ucn2 in CHF$^{4}$ that likely reflect the attendant declines in LAP (and thus reduction in cardiac transmural distending pressures) leading to reduced stimulus for secretion and release. Although plasma concentrations of the natriuretic peptides decreased during Ucn2 treatment (in response to hemodynamic improvement), it should be noted that Ucn2 actually promotes atrial natriuretic peptide and brain natriuretic peptide secretion in cardiomyocytes$^{28}$ (and possibly other organs) and may therefore still enhance the beneficial actions of these peptides locally.

The salutary attenuation of plasma AVP by Ucn2,$^{4}$ seen whether the peptide was administered alone or conjointly with furosemide, is presumably attributable to reduced circulating levels of its secretagogue-angiotensin II$^{29}$ and to improvements in CO (and thus pressure to sino-aortic volume receptors). Overall, these data demonstrate a beneficial effect of Ucn2 adjunct therapy to suppress volume-retaining and vasoconstricting neurohumoral factors in this setting, particularly pertinent in the light of furosemide’s deleterious neurohormonal actions in CHF.

The hemodynamic responses to acute furosemide administration observed in this study are consistent with our earlier reports in experimental ovine CHF,$^{19}$ with reductions in cardiac preload and afterload reflecting reduced plasma volume (evidenced by the relative increase in hematocrit) and direct vascular actions$^{30,31}$ (as judged here by the decrease in CTPR). Cotreatment with Ucn2 produced dramatically greater decreases in LAP, likely a consequence of concomitant improvements in CO, although the lusitropic actions of the peptide,$^{3}$ and possible venodilator actions to reduce circulatory filling pressures,$^{32}$ may also have played a part. The Ucn2-induced augmentation in CO presumably reflects decreases in cardiac afterload (CTPR) and the peptide’s potent inotropic activity,$^{3}$ as suggested by the concurrent

![Figure 3. Mean±SEM hormonal responses to 3-hour infusions of vehicle (C), Ucn2 (50 μg bolus+50 μg/h infusion; #), furosemide (20 mg bolus+3.3 mg/h infusion; @), and Ucn2+furosemide combined (■) in 7 sheep with heart failure.](http://circheartfailure.ahajournals.org/doi/abs/10.1161/CIRCHEARTFAILURE.109.802563)
increase in dP/dt(max). This observation is consistent with reports demonstrating that Ucn2 enhances contractility in isolated cardiomyocytes.33 Although the increase in CO might not be perceived as propitious in terms of possible increased myocardial oxygen demand, the additional actions of the Ucn2 to improve cardiac bioenergetics (through preservation of high-energy phosphate stores)34 and cardiomyocyte intracellular calcium handling,33 dilate coronary arteries,35 and lessen cardiac workload through reductions in peripheral resistance, may counter any adverse consequence. The cardioprotective and antiarrhythmic activities7 demonstrated by the UcnS may further contribute to their beneficial cardiac effects in the setting of CHF. The latter action may be especially germane in this setting because aggressive diuretic therapy carries a danger of arrhythmias subsequent to severe electrolyte and volume depletion.24

Although Ucn2 is a potent vasodilator, acting directly on vascular tone26 (indicated here by significant reductions in CTPR) and indirectly by reducing circulating levels of the vasoconstrictor angiotensin II23 (as assessed by decreases in PRA) and by inhibiting endothelin-1-induced arterial constriction,35 the combination of Ucn2 and furosemide did not result in an additional decrease in MAP above that achieved by furosemide alone. This is likely to be attributed to the sizable improvement in CO induced by Ucn2. From a therapeutic perspective, the combination of Ucn2 and diuretic therapy (producing further declines in peripheral resistance without supplementary reductions in blood pressure) could be appealing in this respect because excessive diuresis bears the potential risk of hypotension and worsening renal function.24

Intriguingly, hematocrit dropped after Ucn2 treatment, despite an active diuresis and associated negative fluid balance in these animals, conceivably pointing to an acute shift of fluid from the extravascular to vascular compartments. It is also possible that the peptide has an effect on red marrow effects are usually seen over days/weeks and months. The Ucn2-induced reduction in hematocrit was mitigated by contemporaneous furosemide administration, presumably reflecting the markedly greater negative fluid balance status demonstrated in this group.

In conclusion, Ucn2 cotreatment with furosemide in an experimental model of CHF-enhanced hemodynamic and renal function and diuretic responsiveness, without additional potassium depletion. Importantly, Ucn2 also reversed furosemide-induced increases in PRA and induced greater decreases in plasma aldosterone and vasopressin. Our findings indicate that adjunct Ucn2 therapy may represent a useful strategy in CHF. Whether this agent will prove effective in diuretic-resistant patients awaits further investigation.

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Disclosures
None.

References
This study demonstrates that acute Ucn2 cotreatment with the loop diuretic furosemide in experimental congestive heart failure produces additional natriuresis and diuresis in conjunction with an increase in glomerular filtration rate (as judged by creatinine clearance) but without added kaliuresis. Importantly, Ucn2 coadministration reversed furosemide-induced stimulation of plasma renin activity and enhanced the inhibitory action of the diuretic on plasma levels of aldosterone and vasopressin. Although furosemide alone had little or no effect on cardiac output, left atrial pressure, and calculated total peripheral resistance, the addition of Ucn2 had sizable beneficial actions on each of these hemodynamic indices. These data demonstrate substantial short-term beneficial effects of Ucn2 on hemodynamics, renal function, and neurohumoral indices when added to loop diuretic therapy in experimental congestive heart failure. Our findings may be of clinical relevance given the risks, actual and potential, associated with loop diuretic therapy (especially during aggressive or chronic therapy), which include a decline in glomerular filtration rate and stimulation of the renin-angiotensin system and aldosterone (with attendant deleterious effects on systemic vascular resistance, sodium, potassium, and magnesium balance, and cardiovascular remodeling). These observations, along with earlier reports of cardioprotective and antiaarrhythmic activities of Ucn2, suggest that studies of more prolonged administration of Ucn2 in both experimental animals and patients with congestive heart failure are warranted.

**CLINICAL PERSPECTIVE**

Urocortin 2 Inhibits Furosemide-Induced Activation of Renin and Enhances Renal Function and Diuretic Responsiveness in Experimental Heart Failure
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