Low plasma renin and reduced renin secretory responses to acute stimuli in conscious COX-2-deficient mice

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Kim SM, Chen L, Mizel D, Huang YG, Briggs JP, Schnermann J. Low plasma renin and reduced renin secretory responses to acute stimuli in conscious COX-2-deficient mice. Am J Physiol Renal Physiol 292: F415–F422, 2007. First published September 5, 2006; doi:10.1152/ajprenal.00317.2006.—In the current experiments, we determined the response of plasma renin concentration (PRC) to acute intraperitoneal administration of furosemide (40 mg/kg), hydralazine (2 mg/kg), isoproterenol (10 mg/kg), candesartan (50 μg), or quinaprilate (50 μg) in conscious wild-type (WT) and cyclooxygenase (COX)-2−/− mice on three different genetic backgrounds (mixed, C57BL/6, 129J). PRC was measured in plasma obtained by tail vein puncture. Basal PRC was significantly lower in COX-2−/− than WT mice independent of genetic background (51, 10, and 17% of WT in mixed, 129J, and C57BL/6). All five acute interventions caused significant increases of PRC in both COX-2+/+ and −/− mice, but the response was consistently less in COX-2−/− mice (e.g., ΔPRC in mg ANG I·ml⁻¹·h⁻¹ caused by furosemide, isoproterenol, hydralazine, quinaprilate, or candesartan 4,699 ± 544, 3,534 ± 957, 2,522 ± 369, 9,453 ± 1,705, 66,455 ± 21,938 in 129J WT, and 201 ± 78, 869 ± 275, 140 ± 71, 902 ± 304, 2,660 ± 954 in 129J COX-2−/−). A low-NaCl diet and enalapril for 1 wk caused a 14-fold elevation of PRC in COX-2−/− mice and was associated with a greatly increased PRC response to acute furosemide (ΔPRC 201 ± 78 before and 15,984 ± 2,397 after low Na/enalapril). As measured by radiotetemetry, blood pressure and heart rate responses to furosemide, hydralazine, isoproterenol, candesartan, or quinaprilate were not different between COX-2 genotypes. In conclusion, chronic absence of COX-2 reduces renin expression, release, and PRC and is associated with a reduced ability to alter PRC during acute stimulation regardless of the nature of the stimulus. COX-2 activity does not appear to be a mandatory and specific requirement for furosemide-stimulated renin secretion.

furosemide; hydralazine; isoproterenol; quinaprilate; candesartan; genetic background; cyclooxygenase-2

INCREASES OR DECREASES OF NaCl concentration in the tubular fluid at the level of the macula densa are followed by inverse changes in the amount of renin released from juxtaglomerular cell stores, an event that is followed by changes in plasma renin concentration (PRC) and ANG II formation rate. Prostaglandins, specifically PGE2 and PGI2, are known regulators of renin release, and they have been implicated in macula densa-mediated regulation of renin release (12). There is considerable support for the notion that PGE2 is generated in macula densa cells through the action of cyclooxygenase (COX)-2 and that it is released when NaCl concentration at the macula densa is acutely reduced (20). In addition to the response of renin release to acute luminal NaCl perturbations, prolonged deviations of NaCl concentration at the macula densa are also believed to cause alterations in renin gene expression and renal renin content. For example, administration of a high-salt diet or chronic infusion of loop diuretics causes changes in renin mRNA and renal renin content that subsequently lead to changes of PRC. It is likely that prostaglandins are also involved in this up- or downregulation of renin expression and the change in plasma renin that follows it. A role for COX-2 in the regulation of renin expression is supported by the markedly reduced levels of renin mRNA in COX-2-deficient mice (23).

The current experiments aimed at confirming the renin-suppressing effect of chronic COX-2 deficiency and exploring whether downregulation of renin expression and PRC may become a limiting factor for the secretory response to subsequent acute stimulation. Our results show that the previously described decrease of renin expression in COX-2-deficient mice is accompanied by a marked reduction in plasma renin and that an acute stimulation of renin release by furosemide, hydralazine, isoproterenol, an angiotensin-converting enzyme inhibitor, or an angiotensin receptor blocker all induce changes in PRC that are much smaller than those observed in wild-type (WT) animals. Thus these studies in COX-2-deficient mice cast some doubt on the notion that COX-2 inhibition interferes specifically with the macula densa pathway of renin secretion. Rather they seem to indicate that chronic COX-2 deficiency causes a reduction in the releasable renin pool and that this affects the acute release response in a nonspecific fashion.

METHODS

Animals. Studies were performed in WT and COX-2 null mutant mice. Mice used had three different genetic backgrounds. Besides animals with a mixed 129J/BL6 background originally generated by Dinchuk et al. (8) and distributed by Jackson Laboratories (Bar Harbor, ME), we used congenic strains on a 129J/Sv and C57BL/6 background generated in our laboratory (24). Littermates from heterozygous breeding pairs were used in each strain as WT controls. At the time of study, mice were between 8 and 12 wk of age. Genotyping was done on tail DNA using PCR as described previously (24). All mice were kept on a standard rodent chow and tap water. Animal care and experimentation were approved and carried out in accordance with National Institutes of Health Principles and Guidelines for the Care and Use of Laboratory Animals.

RNA quantification. PCR reactions (total volume of 10 μl) included cDNA, 900 nM primers, 250 nM probe, and 5 μl of TaqMan MasterMix (Applied Biosystems, Foster City, CA). Total RNA was extracted (RNeasy Mini kit, Qiagen) and treated with DNase I (Invitrogen) at room temperature for 15 min. Reverse transcription was performed using SuperScript II (SuperScript II first-strand synthesis system for RT-PCR, Invitrogen). Renin mRNA levels were

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assessed by real-time PCR using β-actin cDNA (primers and probe from Applied Biosystems) as an internal control. The following primers were used to amplify a 98-bp renin product: sense 5'-CACACTCAAGCAGTCAGGAGCTC (9952–9975), antisense 5'-CAGTGGGTCGTGGGGAGTGC (10613–10595), probe CTACAGTATCCCCACAGGA (9977–10559).

Blood collection and renin determination. Blood was taken from conscious mice by tail vein puncture and collected into a 75-μl hematocrit tube that contained 1 μl 125 mM EDTA in its tip. Red cells and plasma were separated by centrifugation and frozen until used for renin determination. With the use of a fivefold dilution of 2 μl of plasma, renin concentration was measured by radioimmunoassay (Gammacoat, DiaSorin, Stillwater, MN) as the generation of ANG I after addition of excess rat substrate with final plasma dilutions varying between 1:500 and 1:10,000. ANG I generation was determined for a 1-h incubation period at 37°C and expressed as an hourly average. In each assay, background ANG I formation was determined by incubating substrate without plasma for the same time and was subtracted from the plasma containing samples if necessary. In addition, background ANG I levels were determined in a plasma aliquot kept frozen without the addition of substrate until assaying. All plasma samples were stored at −20°C until assay.

Blood collections for PRC measurements were usually made between 10 and 12 AM. In 10 WT and 8 COX-2−/− mice on 129 J background, we compared these morning values of PRC with midnight values of PRC in individual wild-type mice with mixed (1–4) or 129J genetic backgrounds (5–8). Numbers above the abscissa indicate the ratio of highest to lowest values in each animal.

Blood pressure telemetry. Telemetric transmitters were magnetically activated >24 h before implantation. Mice were anesthetized with ketamine and xylazine (90 and 10 mg/kg, respectively), and the left carotid artery was isolated. The telemeter catheter was inserted into the left carotid artery and advanced to reach the aortic arch, and the telemeter body (model TA11PA-C20, Data Sciences International, St. Paul, MN) was placed in a subcutaneous pocket on the right flank (1, 2). One day after surgery, each animal was returned to its home cage with ad libitum food and water for the duration of the study. The telemeter signal was processed using a model RPC-1 receiver, a 20-channel data-exchange matrix, APR-1 ambient pressure monitor, and a Dataquest ART 2.3 acquisition system (Data Sciences International). The system was programmed to acquire data for 10 s every 2 min and to calculate 10-min averages of the mean, systolic, and diastolic blood pressure, pulse pressure, heart rate, and activity. The recording room was maintained at 21–22°C with a 12:12-h light-dark cycle. The implanted telemeter was activated on the morning of day 10, and the mice were left undisturbed for at least 5 h while their blood pressure was recorded. Recordings continued while mice were treated with furosemide, isoproterenol, hydralazine, candesartan, or quinaprilate.

Study protocols. All agents were given by intraperitoneal injection with blood collections made 60 min after the injection. Agents were given in the following doses: 40 mg/kg furosemide (Lasix, Hoechst); 1 mg/kg hydralazine (Sigma); 10 mg/kg isoproterenol; 50 μg/mouse quinaprilate; 50 μg candesartan/mouse. The low-salt diet was a commercial diet containing 0.03% NaCl, and it was given together with enalapril in the drinking water (150 mg/l) providing ~10 mg·kg⁻¹·day⁻¹ of the drug.

Statistics. Data are expressed as means ± SE. Statistical comparisons were done by paired t-test for comparisons of PRC before and after an intervention in the same animals, and by unpaired t-test for comparisons between two groups of animals, for example of different genotypes. ANOVA with repeated measures was used for comparisons of more than two data sets. P values smaller than 0.05 were considered to indicate a significant difference.

RESULTS

Basal PRC. Since in some of our protocols we compared PRC measurements made on different days, we assessed the spontaneous variability of PRC values in individual mice by making repeated measurements in WT mice. Figure 1 summarizes the results from four mice with mixed background in which five blood collections were made over 10 days and from four mice with 129J background in which eight blood collections over 16 days were done. Our data show that PRC values vary more between than within individual mice (mean standard deviation 509 between vs. 321 within). The mean ratio of the highest to the lowest value in each mouse averaged 2.75 suggesting that greater than threefold changes are unlikely to be due to spontaneous variations.

Measurements of basal PRC in three strains of WT and COX-2−/− mice are shown in Fig. 2. As noted earlier, there was no significant difference in basal PRC between strains, although 129J mice possess two renin genes while C57BL/6 have only one (11). Our data indicate that basal PRC is significantly lower in COX-2−/− mice compared with WT independent of genetic background (Fig. 2). Nevertheless, while PRC in COX-2−/− mice on a mixed background was...
As can be seen, furosemide caused significant increases of PRC in WT mice independent of genetic background. Thus, after low Na+/enalapril treatment of COX-2-deficient mice, the increase of PRC caused by furosemide was markedly greater in WT than in COX-2-deficient animals, and there was a correlation between basal PRC and the PCR increment caused by hydralazine (Fig. 5B). The blood pressure reduction and the increase in heart rate caused by hydralazine were comparable in WT and COX-2−/− mice, ~20 mmHg and 50 beats/min (Fig. 4B).

Effect of isoproterenol. The β-adrenergic agonist isoproterenol (10 mg/kg) increased PRC significantly in WT mice as well as in COX-2-deficient mice on all genetic backgrounds (Fig. 6A). As with furosemide and hydralazine, the magnitude of the response to acute β-adrenergic stimulation was markedly smaller in COX-2-deficient than WT mice, and the isoproterenol-induced change in PRC correlated closely with basal PRC values (Fig. 6B; r² of individual data 0.16, slope P = 0.004). The hemodynamic response to acute isoproterenol injections consisted of transient decreases of blood pressure and increases of heart rate, and these changes were comparable between COX-2−/− and WT animals (Fig. 4C).

Effect of candesartan and quinaprilate. Acute administration of the AT₁ receptor blocker candesartan (50 µg) caused marked increases of PRC in WT mice independent of genetic background (33-fold in 129J and 21-fold in mixed background mice). There was also a significant increase of PRC in COX-2−/− mice (11.8-fold in 129J and 12.4-fold in mixed background mice). However, the absolute change of PRC caused by candesartan was markedly reduced in COX-2−/− mice (Fig. 7A, top). The change in PRC caused by candesartan correlated with basal PRC (Fig. 7A, bottom; r² of individual data 0.43, slope P < 0.0001). Candesartan did not significantly alter arterial blood pressure in either WT or COX-2−/− mice, whereas it increased heart rates in both genotypes (Fig. 8A).

Qualitatively similar results were obtained with the ACE inhibitor quinaprilate (50 µg). The agent caused a large increase in PRC in WT animals (12.2-fold in 129J and 13.1-fold in mixed background mice). PRC also increased in COX-2−/− mice, but the absolute elevation of PRC was markedly less in the COX-2-deficient than the WT animals (Fig. 7B, top). Mean changes on PRC induced by quinaprilate correlated with basal PRC (Fig. 7B, bottom; r² of individual data 0.17, slope P = 0.02). Quinaprilate caused a blood pressure reduction of ~25–30 mmHg in both WT and COX-2−/− mice (129J background) and a reflex acceleration of heart rates (Fig. 8B).

Effect of low-salt diet and enalapril. To study the effect of prior upregulation of renin on the acute response to furosemide, mice were treated for 1 wk with a low-salt diet in combination with enalapril (10 mg·kg⁻¹·day⁻¹). As shown in Fig. 9, renal renin mRNA following treatment increased to 303.3 ± 62% of control in WT (P < 0.001; n = 7) and to 56.6 ± 10% in COX-2−/− mice (P < 0.01; n = 6). Concomitantly (Fig. 10) PRC (ng ANG I·ml⁻¹·h⁻¹) increased from 1,585 ± 223 to 31,323 ± 5,570 in WT (n = 13; P < 0.001) and from 229 ± 85 to 3,314 ± 949 in COX-2−/− mice (n = 11; P < 0.001; 129J background). The acute administration of furosemide (40 mg/kg ip) increased PRC to 72,741 ± 8,474 in WT (P = 0.002) and to 19,298 ± 2,920 in COX-2−/− mice (P < 0.001). Thus, after low Na/+enalapril treatment of COX-2-null mice, the increase of PRC caused by furosemide was greater than that found in intact control animals (Fig. 10).
COX-2 appears to be an important determinant of basal renin expression, renin release, and PRC. However, when renin expression and PRC are normalized in the absence of COX-2, full responsiveness of PRC to acute furosemide administration is reestablished.

DISCUSSION

The purpose of the present study was to further explore the relationship between COX-2 and the release of renin from the renal juxtaglomerular apparatus. The main result is the observation that genetic COX-2 deficiency is associated with a significant reduction of PRC and with a consistent decrease in the absolute magnitude of the change of PRC caused by a number of known acute stimulators of renin secretion. We suggest that these two effects reflect a cause-and-effect relationship, i.e., that a reduction in renal renin expression and PRC caused by the chronic absence of a stimulatory COX-2 product leads to a reduction in the amount of renin that can be released in response to acute stimulation and that this attenuation is independent of the nature of the stimulus.

In the present experiments, we used genetically COX-2-deficient mice to ensure chronic, complete, and selective absence of COX-2 enzyme activity. The advantages of this approach must be weighed against the known renal abnormalities, most notably in the postnatal development of superficial glomeruli, that are a characteristic feature of genetic COX-2 deficiency (8, 18). To minimize the impact of these changes, we used mice between 8 and 12 wk where overall renal function is largely unaltered. Furthermore, we used three mouse strains that differ in their genetic background and have been shown to vary in their susceptibility to developing renal failure (24). Since the regulation of the renin-angiotensin system was qualitatively the same between these strains, we assume that effects secondary to glomerular damage had no major impact on the outcome of our study. Thus the marked reduction of renin expression and PRC in COX-2-deficient mice with different genetic backgrounds indicates that COX-2 plays a critical role in maintaining normal renin levels. These observations are in agreement with earlier measurements from our laboratory demonstrating markedly reduced levels of renal renin mRNA and of glomerular renin protein and enzyme activity (23). Furthermore, we demonstrated reduced PRC values in COX-2−/− mice on two different genetic backgrounds, C57BL/6 and BALB/c (19). These previous and our present observations are somewhat at variance with another study in COX-2−/− mice in which neither renal renin mRNA, renal renin activity, nor plasma renin activity were significantly reduced compared with WT animals (7). It is noteworthy that these studies were done in COX-2−/− mice on a mixed genetic background since the present studies show that the reduction in PRC for unknown reasons is less pronounced in mixed background animals compared with COX-2−/− mice on a C57BL/6 or 129J background. The COX-2 inhibitor SC-58125 has been found to reduce plasma renin activity and
renal renin mRNA levels in low salt-fed and renal hypertensive mice (9). On the other hand, an up to 3-wk treatment with the COX-2 inhibitor rofecoxib did not alter renin mRNA or plasma renin activity in rats (14, 15). A striking difference between these and the present studies is the marked upregulation of COX-2 expression in the rofecoxib-treated rats (14). It is possible that the pharmacological approach may not abolish local PG production, either because of accessibility problems or because the upregulated COX-2 expression enhances enzyme capacity that, in part, compensates for reduced enzyme activity. From our observations in genetically COX-2-deficient mice and in JG cells isolated from these mice, we conclude that tonic COX-2 activity is required to maintain renin expression in juxtaglomerular cells at levels high enough to sustain normal rates of renin release and normal PRCs.

Acute administration of furosemide caused a significant increase of PRC in WT mice as shown previously (3, 16). Furosemide is thought to elicit its renin stimulatory action by elimination of a renin inhibitory pathway that is dependent on activation of NKCC2 in macula densa cells. Previous results have suggested that the generation of a prostaglandin is a downstream event that follows inhibition of NKCC2 by low NaCl or loop diuretics and that an acute increase of PG formation is required for macula densa-dependent stimulation of renin secretion (10, 21). In fact, a low luminal NaCl concentration has recently been shown to cause release of PGE2 from macula densa cells isolated from the kidneys of rabbits maintained on a low-NaCl intake (20). Our observation that the absolute magnitude of the increase in PRC following furosemide administration was markedly reduced in COX-2−/− mice would appear to be in general agreement with the notion that COX-2-dependent PG formation is required for macula densa-mediated renin release. However, previous results from our laboratory have shown that COX-2 deficiency is also associated with a reduction of renin expression (23). Conceivably therefore, the reduced renin release following acute stimulation by furosemide in COX-2−/− mice may be a reflection of a chronic reduction in the releasable pool of renin rather than an indication of a change in the signaling pathway activated by acute furosemide administration.

To further investigate this issue, we compared the effect of other acute stimuli of renin release on PRC in WT and COX-2−/− animals. Hydralazine is a vasodilating agent that elicits renin secretion mostly by blood pressure reduction and subsequent unloading of the renal baroreceptor. Previous evidence indicated that the baroreceptor mechanism of renin release is not obligatorily dependent on an intact prostaglandin formation (22). Nevertheless, our current data show that the increase in PRC following acute hydralazine administration was significantly reduced in COX-2−/− compared with WT mice. Application of isoproterenol was used as another means to acutely stimulate renin release. Renin stimulation by β-ad-

Fig. 5. A: mean increase of PRC over basal caused by hydralazine (1 mg/kg ip) in COX-2+/+ and COX-2−/− mice with mixed (n = 9 and n = 6), C57BL/6 (n = 5 and n = 7), or 129J backgrounds (n = 13 and n = 9). **P < 0.001, #P < 0.05 (significance of change, paired test). B: relationship between mean hydralazine-induced PRC increase and mean basal PRC in COX-2+/+ and COX-2−/− mice and linear regression line.

Fig. 6. A: mean increase of PRC over basal caused by isoproterenol (10 mg/kg ip) in COX-2+/+ and COX-2−/− mice with mixed (n = 9 and n = 6), C57BL/6 (n = 5 and n = 7), or 129J backgrounds (n = 13 and n = 9). **P < 0.001, *P < 0.05 (significance of change, paired test). B: relationship between mean isoproterenol-induced PRC increase and mean basal PRC in COX-2+/+ and COX-2−/− mice and linear regression line.
energic activation has been shown to be independent of prostaglandin synthesis (13, 17). As with furosemide and hydralazine, however, the rise of PRC following isoproterenol was markedly diminished in COX-2-deficient mice. The stereotypic reduction of renin secretion in COX-2-deficient mice to stimuli which are unlikely to share a specific mechanistic PG dependency is most easily explainable by the commonality of a reduction in the amount of renin available for acute release. If this notion is in fact correct, one may expect other conditions of low renin expression to likewise lead to a reduced secretory

![Image](image1.png)

**Fig. 7.** A: mean increase of PRC over basal caused by candesartan (50 μg/mouse) in COX-2+/+ and COX-2−/− mice with mixed (n = 9 and n = 6) or 129J backgrounds (n = 9 and n = 7; **P < 0.001, *P < 0.05, paired test), relationship between mean candesartan-induced PRC increase and mean basal PRC, and linear regression line. B: mean increase of PRC over basal caused by quinaprilate (50 μg/mouse) in COX-2+/+ and COX-2−/− mice with mixed (n = 9 and n = 6) or 129J backgrounds (n = 9 and n = 7; **P < 0.001, *P < 0.05, paired test), relationship between mean quinaprilate-induced PRC increase and mean basal PRC, and linear regression line.

![Image](image2.png)

**Fig. 8.** Telemetric recordings of MAP (±SE) and HR (±SE) in COX-2+/+ and COX-2−/− mice on 129J genetic background before and after the injection of candesartan (A) or quinaprilate (B).

![Image](image3.png)

**Fig. 9.** A: renal renin mRNA of COX-2+/+ and COX-2−/− on 129J background under basal conditions (n = 4 and n = 7 for COX-2+/+ and COX-2−/−, respectively) and after a 1-wk treatment with a low-NaCl diet and enalapril (n = 5 and n = 6 for COX-2+/+ and COX-2−/−, respectively) as determined by quantitative PCR and expressed as percent of COX-2+/+ control. Significances are given for comparisons between untreated control and treated animals. B: mean PRC (±SE) in COX-2+/+ and COX-2−/− mice with 129J genetic backgrounds. P values indicate significance levels between wild-type and COX-2-deficient mice (unpaired t-test); numbers inside bars indicate number of individual mice in each group.
response to acute stimulation. In fact, previous results from our laboratory showed that β1/β2-adrenergic receptor-deficient mice also have markedly reduced PRC values and that the PRC response to both furosemide and hydralazine was clearly reduced in these animals compared with WT controls (16).

Further emphasizing the importance of basal levels of renin for the magnitude of the acute secretory response are our observations with angiotensin receptor blockade and ACE inhibition. Acute administration of either the AT1 or the ACE inhibitor causes a drastic elevation of PRC in WT mice that most likely reflects stimulation of renin secretion. We cannot fully exclude the possibility that these agents may affect renin gene expression within the time span of these studies, but in general upregulation of renal renin content is relatively slow, taking place over periods of hours rather than minutes (5). Candesartan and quinaprilate also elevated PRC in COX-2−/− mice, but as with the other acute stimuli the magnitude of the PRC change was substantially reduced. COX-2 deficiency may interfere in some specific way with the mechanism of action of AT1 or ACE inhibitors, but on the background of all of our data it seems more likely that this reduction is another documentation of the effect of a chronically downregulated renal renin content on the acute renin secretory responsiveness.

This notion is supported by the effect of an upregulation of PRC achieved by treating COX-2-deficient mice with enalapril and low-NaCl intake for 1 wk. As expected, this treatment augmented PRC of WT mice to supranormal levels, while it increased PRC values in COX-2−/− mice to values that exceed basal WT levels while remaining well below those of enalapril-treated WT mice. These results are in agreement with previous studies showing a reduced effectiveness of chronic ACEI administration in upregulating renal renin expression in COX-2−/− mice or in the chronic presence of a COX-2-specific inhibitor (6, 7). Upregulation of PRC in COX-2−/− mice by low NaCl/enalapril treatment was associated with normalization of the PRC elevating effect of acute furosemide administration indicating that under these conditions activation of the macula densa pathway can stimulate renin release without COX-2. Thus we conclude that COX-2 deficiency reduces the magnitude of the PRC change following furosemide only when basal PRC levels are suppressed but not when they are upregulated before drug application. This finding suggests that COX-2 is not absolutely necessary for furosemide-stimulated renin secretion.

Previous studies in COX-2−/− mice with a mixed genetic background have failed to show significant increases of renal renin mRNA expression, renal renin content, or plasma renin activity in response to chronic ACE inhibition with captopril (7). Other studies showed that the stimulation of renin by chronic captopril was blunted by the COX-2 inhibitor SC58236 (6). Taken together, these studies seemed to indicate that COX-2 is an important regulator of renin and that termination of ANG II-dependent suppression of COX-2 mediates the increased renin production in response to inhibition of ANG II production or action. However, in contrast to these studies the current results demonstrate that chronic ACEI in combination with low-Na intake markedly elevates PRC (from 229 to 3,314 ng ANG I·ml−1·h−1), an effect that in this genetic model of COX-2 deficiency cannot be mediated by interruption of ANG II feedback suppression of COX-2. We suspect rather that the chronic stimulatory effects of COX-2 and the inhibitory effects of ANG II are independent and additive determinants of renin synthesis and release. Thus, even in the complete absence of COX-2, removal of ANG II inhibition by ACEI is still capable of increasing renin, although not to the extent achievable with an intact COX-2 influence.

In summary, the markedly reduced levels of plasma renin and the reduction in renin gene expression in COX-2-deficient mice indicate that a functional COX-2 is critical for maintaining a normal size of the releasable renin pool. Decreased expression of renin in COX-2-deficient mice is associated with a marked reduction of the response of PRC to acute administration of furosemide, isoproterenol, hydralazine, quinaprilate, or candesartan suggesting that the level of renin expression is a nonspecific determinant of the magnitude of the acute release response. Upregulation of renin expression and normalization of PRC in COX-2-deficient mice by chronic low NaCl/enalapril pretreatment restored PRC responses to furosemide. Thus, under these experimental conditions, COX-2 does not appear to be required for furosemide to elicit an increase of renin release of normal magnitude.

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