Angiotensin II Decreases the Renal MRI Blood Oxygenation Level–Dependent Signal

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Abstract—Acute experimental reduction of renal blood flow decreases the renal blood oxygenation level–dependent (BOLD) MRI signal in animals. Angiotensin II also reduces renal blood flow, but the ability of BOLD MRI to dynamically detect this response has not yet been investigated in humans. Six healthy male volunteers underwent an individual dose-finding study to identify the intravenous doses of angiotensin II, norepinephrine, and sodium nitroprusside necessary to induce a 15-mm Hg peak mean arterial blood pressure change. MRI studies followed within 3 weeks, when angiotensin II (8.8±1.4 ng/kg), norepinephrine (52±12 ng/kg), and sodium nitroprusside (2.0±0.3 μg/kg) were given twice in an unblocked, randomized sequence while imaging experiments were performed on a 1.5-T Siemens Sonata. A multiecho echo-planar imaging sequence was used to acquire T2* maps with a temporal resolution of 1 respiratory cycle. Averaged over a renal cortex dominated region of interest, angiotensin II caused a shortening of T2* between 6% and 10%. Sodium nitroprusside and norepinephrine, although of equal potency concerning blood pressure responses, did not alter the renal BOLD signal. The renal BOLD response to angiotensin II appeared with short onset latency (as early as 10 seconds after peripheral intravenous angiotensin II bolus administration) suggesting that this response is a consequence of altered perfusion rather than increased renal oxygen consumption. The methods described here are suitable to assess renal responsiveness to angiotensin II and may, thus, be of great value in human hypertension research. (Hypertension. 2006;47:1062-1066.)

Key Words: kidney ■ magnetic resonance imaging ■ blood pressure ■ angiotensin II ■ norepinephrine ■ nitroprusside

Inappropriate activation of the renin–angiotensin–aldosterone–system may result in the development of arterial hypertension.1 Chronic infusions of subpressor doses of angiotensin II (A2) induce long-term blood pressure (BP) increases.2 This effect is likely mediated by the kidneys.3,4 A2 affects a variety of kidney functions potentially influencing long-term BP control, such as renal handling of sodium5 and renal hemodynamics.6 Clearance techniques may be applied in humans to study such effects but are time consuming. Alternative noninvasive techniques are warranted. It was demonstrated recently in animals that acute experimental reduction of renal perfusion pressure, with subsequent reduction of renal blood flow, diminishes the renal blood oxygenation level–dependent (BOLD) MRI signal.7,8

BOLD MRI exploits the fact that the different magnetic susceptibilities of oxygenated and deoxygenated hemoglobin result in locally different effective transversal relaxation rates. Therefore, changes in tissue oxygenation can be detected by BOLD MRI via altered concentrations of deoxyhemoglobin. Because A2 reduces renal blood flow,6,9 we hypothesized that A2 reduces the renal BOLD MRI signal, as well. Norepinephrine (NE) and sodium nitroprusside (SNP) are potent vasoactive substances with excellent dose-response characteristics. They may be administered in individually adjusted doses as to affect arterial BP and renal perfusion pressure at the same extend as does A2. Previous research indicated NE10,11 and SNP12,13 to have little or no impact on renal perfusion because of renal autoregulation. Thus, we hypothesized that NE and SNP also have no effect on the BOLD MRI signal and used these substances as control interventions.

Methods

Six healthy male volunteers (mean age, 26; range, 22 to 29 years; body weight, 71.2; range, 64 to 76 kg) participated. They had normal BP, normal values during physical examination, electrocardiography, routine blood chemistry, and hematology; normal urine sediment; and no microalbuminuria. None of the subjects took any medication. They were asked to avoid physical exercise (eg, riding a bicycle) and to refrain from drinking caffeine-containing beverages on the morning of the study days. Volunteers provided informed consent before studies were performed.

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consent. The research protocol was approved by the local ethics committee.

Subjects were examined on 2 occasions, separated by 3 weeks. The first examination was to individually identify bolus A2, NE, and SNP doses evoking a peak 15-mm Hg mean arterial BP change (dose-finding day). The second examination was to apply the identified doses during renal MRI (magnetic resonance [MR] study day). Unless impossible or otherwise stated, procedures and instructions were similar on both study days, as was body position.

**Dose-Finding Day**
All of the volunteers underwent an individual dose-finding study. They entered the hospital at ~10:00 AM, and a small venous catheter was attached intravenously into a prominent cubital vein for bolus drug infusion. Volunteers remained in a recumbent position during the following manipulations. A2 was purchased from Clinalfa AG, NE from Sintetica S.A., and SNP (Nipruss) from Schwarz Pharma. Beat-to-beat heart rate (HR) was assessed by conventional single-lead ECG. Continuous BP was assessed noninvasively with Finapres 2300 system (Ohmeda Inc, Englewood). Data were recorded for 5 minutes (1 minute before and 4 minutes after infusion). All of the vasoactive substances were titrated as to induce a 15-mm Hg peak mean arterial BP change. Reflexive HR changes were assessed, and finger tip perfusion was determined by laser Doppler flowmetry using a Periflux device (PF3, Perimed). After doses were once identified (A2, 8.8±1.4 ng/kg; NE, 52±12 ng/kg; SNP, 2.0±0.3 μg/kg), they were repeated so as to guarantee reproducibility of the mean arterial BP responses within ±3 mm Hg margins.

**MR Study Day**
One MRI experiment consisted of 6 unblocked randomized infusion sessions, each lasting 5 minutes, separated by 10 minutes of washout. All 3 of the vasoactive substances were given twice, and responses were averaged individually and per drug. Dosages were as identified during dose finding. MRI experiments were performed at 1.5 T on a Sonata system (Siemens Medical Solution) using 2 elements of the spine array for signal reception. For 2 reasons, a fast single-shot multiecho echo-planar imaging sequence acquiring 3 echo images after a single radio frequency excitation was chosen for BOLD imaging: (1) it allows us to obtain a sufficient temporal resolution in the dynamic imaging study; and (2) it avoids the application of breath holds, which were considered not to be applicable continuously over the long experimental time of ~5 minutes. Sequence start was triggered by a respiratory signal so as to avoid motion artifacts related to respiration. The imaging sequence had the following parameters: matrix: 128×88; 4 slices; field of view, 30×21 cm²; nominal in-plane resolution, 2.3×2.3 mm²; slice thickness, 5 mm; 3 echo times, 40, 108, and 177 ms; flip angle, 90°; and readout bandwidth, 2604 Hz/pix. Temporal resolution was given by the respiration interval, that is, ~4 s. T2* maps were calculated using a monoexponential model and averaged over a renal cortex–dominated region of interest (ROI). Sagittal views of the right kidney were acquired.

For technical reasons, Finapres cannot be applied in the MR scanner. Thus, it was not possible to assess noninvasive continuous BP while injecting the vasoactive substances during MRI. However, HR was measured by a pulse sensor so as to compare drug-induced HR changes on both study days. As during the dose finding studies, there was 1 minute of baseline scanning before drug administration, which resulted in ~15 baseline images, depending on the respiration interval. MR acquisition was continued for 4 minutes after drug administration.

**Data Analyses**
Variable respiratory cycles lead to image acquisition at different time points for individual subjects. Therefore, measured T2* time courses were interpolated onto a 10-s time scale previous averaging. Differences to the 1-minute baseline period before drug infusion were calculated and are illustrated. For BP and laser Doppler flowmetry, peak drug effects occurred between 60 and 90 s; for HR, peak effects differed for drugs (A2, 30 to 50 s; NE, 80 to 100 s; SNP, 50 to 80 s). The peak T2* renal response to A2 occurred between 30 and 50 s. The above intervals served to average response data and analyze statistical significances with paired t tests at the (2-sided) 5% level. All of the calculations were performed with SAS on a Win XP platform (Version 9, SAS Institute Inc). Means and SDs are provided in text.

**Results**
There were no adverse events. Figure 1 illustrates BP changes and indicates that dose finding was successful. A2 increased BP by 15.0 (3.5) mm Hg (P=0.0001). The NE effect was somewhat (albeit nonsignificantly) smaller, BP increased by 11.8 (3.9) mm Hg (P=0.0008). SNP decreased BP by 12.4 (2.3) mm Hg (P=0.0001). HR decreased reflectively during A2 infusion by 6.2 (3.8) bpm (P=0.01), during NE infusion by 5.2 (2.4) bpm (P=0.003), and increased during SNP infusion by 14.8 (5.2) bpm (P=0.0009). Finger tip skin blood flow responses were increased by SNP (P=0.0001) and decreased by A2 (P=0.003) and NE (P=0.006). Figure 2 indicates that HR changes induced by the vasoactive substances during the MRI study and during the dose-finding study were similar, suggesting comparable underlying BP changes during dose finding and during MR study. An image with the shortest acquired echo time (41 ms), as well as a calculated T2* map and the location of the ROI used for evaluation, are displayed in Figure 3. T2* values measured during the administration of the vasoactive substances are shown in Figures 4 and 5. Average renal cortex baseline T2* was 78.8 (1.4) ms. A2 shortened T2* by 8.2 (1.5) % (P=0.005), whereas neither SNP nor NE lead to significant changes in the transverse relaxation time. For all of the drugs, a control ROI in muscle tissue did not show T2* changes. The calculated mean T2* value in the muscle tissue was 55.7 (5.4) ms and of lower reliability, because the transverse relaxation
time of the psoas muscle at baseline was short compared with the acquired echo times. To overcome this limitation, the first echo images (echo time, 40 ms) of our multiecho sequence were analyzed. Changes in muscle T2* would influence signal intensities in susceptibility-weighted images. With respect to the measured T2* of psoas muscle tissue, the first echo images acquired with an echo time of 40 ms were already strongly susceptibility weighted. No significant signal changes were found in psoas muscle after administration of the vasoactive substances. This finding supports the absence of an effect of the applied drugs on muscle T2* and, therefore, muscle oxygenation.

Discussion

The main new finding of this study is that intravenously administered A2 specifically reduces the renal BOLD signal. This effect is consistent with the well-established property of A2 to diminish renal perfusion6,9 and, therefore, renal oxygenation. The A2-induced decrease of the renal BOLD signal is specific in 2 ways. First, other vasoactive substances, such as NE and SNP, although of equal potency concerning BP changes, did not alter the renal BOLD signal. Second, abdominal muscle control tissue (psoas) did not respond to A2.

Relative to their mass and metabolic capacity, the kidneys are highly perfused. They receive ≈20% of cardiac output, although they represent <2% of the total body weight, and renal oxygen extraction is moderate.15 Hence, it can be assumed that rapid transitions of renal oxygenation (ie, BOLD signal) rather reflect altered blood supply than changing metabolic needs. Indeed, the onset latency of the renal BOLD response to A2 was as short as 10 s after peripheral intravenous bolus A2 administration, clearly faster than the arterial BP response, and, thus, seems to be related to reduced renal perfusion rather than metabolic adjustments.

Renal blood flow is autoregulated over a wide range of renal perfusion pressure, and autoregulation prevents changes in renal blood flow16,17 despite changing renal perfusion pressure. Autoregulation is responsible for almost constant renal blood flow in the presence of SNP-induced decrease of arterial BP and NE-induced increase of arterial BP. A2 is known to override renal autoregulation.6 A2 reduces renal
blood flow in the presence of increased renal perfusion pressure. Currently, renal autoregulation is not fully understood; myogenic and metabolic mechanisms are involved, as is tubuloglomerular feedback. Skin blood flow is poorly autoregulated, and A2 and NE caused a similar reduction in finger tip skin blood flow. In contrast, perfusion of striated muscles is well autoregulated. But unlike in the kidneys, it is not known whether any of the vasoactive substances tested in this study interfere with striated muscle autoregulation of blood supply. BOLD signals may be assessed in striated muscles. However, psoas muscle did not show any drug-induced BOLD signal changes, underpinning the specific nature of the renocortical BOLD response to A2.

All of the vasoactive substances induced the expected HR responses, which were the result of intact baroreflex buffering of BP changes. However, in comparison with NE infusion, HR tended to decelerate early after A2 administration. This difference remains obvious even when taking into account the faster A2-induced pressor response and may reflect agonistic properties of NE on cardiac β-adrenergic receptors.

Several limitations of this study have to be considered. BP was measured by the indirect Finapres system. This system
allows for beat-to-beat assessment of BP changes. Accuracy of this system may be limited under conditions of strong vasoconstriction.20 However, there is evidence that such a bias would influence systolic or diastolic BP readings but not mean arterial pressure.21 Furthermore, this bias would be present during both A2 and NE infusions. Finapres cannot work in the MR scanner. Therefore, we were not able to document whether drug-induced BP reactions in the scanner were identical to the ones assessed during dose finding. However, because drug-induced reflexive HR changes during dose finding and MR study were identical (see Figure 2), it is likely that underlying BP changes were identical as well. Missing drug effects on the psoas muscle BOLD signal may be related to methodological problems, because the transverse relaxation time was rather short at baseline, suggesting limited reliability of the calculated T2* values. Nevertheless, examination of the T2*-weighted first echo images supported the finding of the absence of drug effects. Furthermore, a recent study indicated muscle BOLD signal changes after SNP administration.22 However, methodological differences, most likely the more potent method of intraarterial drug delivery, may explain this discrepancy. The small number of subjects who took part in the study might be seen as a limitation, however, all 6 of the subjects consistently showed the specific responsiveness of kidney oxygenation to A2 administration (see Figure 5).

Perspectives

Data provided in this study are the first to indicate the BOLD-measurable effect of A2 on the human kidney. The method described here is suitable to assess aspects of renal responsiveness to A2 and may, thus, be of great value in human hypertension research. It offers the potential to assess fast and transient components of A2-induced renal responsiveness, and, in perspective, to separate renocortical and renomedullary effects.

References

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