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ORIGINAL RESEARCH

In Patients With Hypertension, the Cortical Perfusion Index Measured by Contrast Enhanced Ultrasound Reveals a Sexual Dimorphism

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BACKGROUND: Arterial hypertension is characterized by microvasculature changes and reduced tissue perfusion. It is unknown whether a sexual dimorphism of renal microcirculation is present in patients with arterial hypertension. We aimed to compare the cortical microcirculation in women and men with hypertension and to evaluate their response to sympathetic stimulation with a cold pressor test (CPT).

METHODS: The cortical perfusion index (PI) measured by contrast-enhanced ultrasonography was used as a proxy of renal microcirculation. We measured the PI at rest and during a 2-minute CPT in patients with arterial hypertension. We used a linear mixed-model analysis to study the effect of sex, the CPT, and their interaction on the PI.

RESULTS: Thirty-two participants were included (34% women). Age and body mass index were similar in both groups. Median PI was higher in women (2344 [interquartile range, 1553–2685 arbitrary units]) than men (1285 [interquartile range, 591–1741 arbitrary units], P=0.009). The CPT increased the PI in both groups; however, the magnitude of the response to CPT was similar in both sexes.

CONCLUSIONS: Women with arterial hypertension have a higher cortical microperfusion than men, while their microvascular reactivity to a CPT appears to be similar. This sexual dimorphism may have an impact on renal function decline, which needs further investigation.

Key Words: contrast-enhanced ultrasound ■ hypertension ■ kidney ■ microcirculation ■ perfusion index ■ sex differences

Sex differences exist in the epidemiology and physiopathology of arterial hypertension (AH).[1] Indeed, compared with men, young women are relatively protected against the development of AH. This is generally explained by differences in lifestyle and sex hormones, which are known to be involved in the renal hemodynamics and the autonomic control of blood pressure (BP).[2,3] Similar observations have been reported in animal studies.[4,5] Moreover, several animal studies suggest that this sexual dimorphism also impacts microvascular function, by affecting blood flow, capillary barrier function, and inflammation.[6,7]

In the kidneys, anatomic and functional changes in the microcirculation may lead to impaired adaptation to vascular stress and insufficient tissue perfusion and lead to the development and progression of chronic

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CLINICAL PERSPECTIVE

What Is New?

- This study examines previously unexplored sex differences of the renal microcirculation of participants with hypertension using contrast-enhanced ultrasonography.
- Renal cortical perfusion expressed as perfusion index is higher in women with hypertension compared with men with hypertension, whereas the renal perfusion index response to a cold pressor test is similar in both sexes.

What Are the Clinical Implications?

- From a diagnostic standpoint, implementing bedside techniques as contrast-enhanced ultrasonography to evaluate renal microcirculation may lead to a more individualized and sex-driven clinical management of arterial hypertension and cardiovascular disease; in addition, evaluation of microcirculation and detection of any sex differences in the kidney microvasculature in other disease states such as diabetes and chronic kidney disease, in which the relevance of renal hemodynamics is of high importance, could help us gain a more comprehensive anatomic and functional picture of the female and male renal microvasculature.

Nonstandard Abbreviations and Acronyms

AH	arterial hypertension
CPT	cold pressor test
PAC	plasma aldosterone concentration
PI	perfusion index
rBV	relative blood volume
RRI	renal resistive index

kidney disease (CKD).[8,9] Thus, a deeper understanding of microvasculature's architecture and functionality is crucial. However, in the absence of imaging modalities that enable high-resolution and non-invasive monitoring of kidney microvasculature in humans, current data mainly come from biopsy and animal studies.[9]

Contrast-enhanced ultrasound (CEUS) associates conventional ultrasonography with the intravenous use of non-nephrotoxic microbubble-based contrast agents that remain intravascular. Thus, CEUS can depict renal vessels as small as 40 μm, and quantify renal microcirculation in different clinical settings and detect changes during physiological or pharmacological stress as previously shown by others and by our group.[10–13]

We recently demonstrated with CEUS that patients with AH have a lower cortical perfusion and a lower vascular response to sympathetic stress than healthy controls.[11] We also reported with CEUS that healthy women have a higher cortical perfusion than men.[14] However, whether there are sex differences in renal microperfusion in people with AH is, to the best of our knowledge, unknown. Therefore, the goal of our study was to compare the cortical microcirculation as assessed by CEUS in the resting state and during a sympathetic stress test in women and men with AH.

METHODS

The data that support the findings of this study are available from the corresponding author upon request.

Study Population

Participants with hypertension were recruited in the outpatient clinic of the Service of Nephrology and Hypertension of the Lausanne University Hospital. Participants could be included if they were aged >18 years, if they had a 24-hour ambulatory BP ≥130/80 mm Hg or treated AH. Participants with prior allergy to ultrasound contrast agent, ongoing pregnancy, acute or chronic disease other than AH affecting renal function, lower-limb sensitive neuropathy, taking drugs affecting renal hemodynamics or antihypertensive treatment other than calcium channel blockers during the washout period, or with office BP >180/100 mm Hg during the washout period were excluded.

Study Design and Settings

This study was a post hoc analysis of a single-center prospective, controlled (patients with hypertension versus patients without hypertension) study evaluating the effect of a CPT on brain and renal function (NCT03473275). In the present study, the PI measured by CEUS in patients with hypertension was the primary outcome. The study was approved by the local ethics committee "Commission cantonale d'éthique de la recherche sur l'être humain" and conducted according to the Declaration of Helsinki and local regulatory requirements. All subjects provided written informed consent before inclusion.

All participants underwent a screening visit. Antihypertensive treatment was standardized before the study visit as follows: no antihypertensive drugs for patients with moderate hypertension (BP <160/100 mm Hg) and amlodipine 5 to 10 mg only for patients with hypertension with more severe AH.

Calcium channel blockers were stopped at least 48 hours before the visit day with the exception of 1 woman and 1 man who took their last dose in the morning before the study day. All other drugs were withheld 10 days before study days.

All participants were instructed to avoid smoking, alcohol intake for 48 hours before the study day, strenuous physical activity 24 hours before the study day, and consumption of caffeine-containing products and energy drinks from 6:00 PM the day before the study. On the study day, fasting participants arrived at the investigation center between 8:00 and 10:00 AM. A venous catheter was inserted in 1 arm to allow intravenous administration of the contrast agent (Sonovue; Bracco International B.V., Milan, Italy) and blood sampling. Estimated glomerular filtration rate was calculated using the Chronic Kidney Disease Epidemiology Collaboration equation.

Systemic Hemodynamic Variables

The pulsatile hemodynamics were recorded using the Mobil-O- Graph (IEM, Stolberg, Germany) in the supine position after 15 minutes of rest.[15,16] At least 5 consecutive measurements of ambulatory brachial BP, separated by 2-minute intervals, were taken in each participant. BP and heart rate were measured continuously using the Finapres NOVA (Finapres Medical Systems, Enschede, The Netherlands). After 5 minutes of signal stabilization, the finger BP was calibrated with a brachial oscillometric BP measurement. This calibration was repeated at least 2 minutes before each CPT.

The baseline systemic hemodynamic variables (systolic BP, diastolic BP, mean MP, and heart rate) were calculated as the average of 2 minutes of continuous recording before the CPT. During the CPT, the systemic hemodynamic variables were calculated as the mean of continuous recordings (up to 2 minutes). Recovery systemic hemodynamic variables were calculated as the mean of the last 60 seconds of the 2-minute recovery period after the CPT.

Hormones

Blood samples were taken before CPT, after 30 seconds, after 2 minutes of CPT, and 2 minutes after the end of CPT. Norepinephrine, epinephrine, and neuropeptide Y 1-36 were determined using a liquid chromatography–tandem mass spectrometry method as reported previously.[17,18] Plasma renin activity and plasma aldosterone concentration (PAC) were determined by a radioimmunoassay kit for the quantitative determination of angiotensin I in human plasma (REN-CT2; Cis-bio Bioassays, Codolet, France) and by a commercial radioimmunoassay kit (Aldo-Riact; CIS Bio International, Yvette, Cedex, Saclay, France), respectively.

Cold Pressor Test

A modified CPT was applied as previously described.[11,12] Both feet were placed in a footbath filled with water at body temperature (35 °C–37 °C, sham) for baseline measurements. The water was replaced by ice-cold water (2 °C–4 °C) for 2 minutes during which renal ultrasound and systemic hemodynamic measurements were repeated. Thereafter, the ice-cold water was substituted by water at body temperature (recovery phase). This procedure was conducted twice with at least 10-minute intervals between the 2 phases: (1) Doppler ultrasound for renal resistive index (RRI) determination and (2) CEUS for PI determination (Figure 1).

Renal Ultrasound Parameters

All subjects underwent a renal Doppler ultrasound followed by CEUS, using a Samsung RS80A device as previously described.[11,12] The Doppler mode was applied to select the segmental artery with the easiest accessibility and highest quality of Doppler waveforms for the measurement of the RRI. For this reason, in almost all participants, the right kidney was chosen. At least 4 measurements of RRI were obtained in

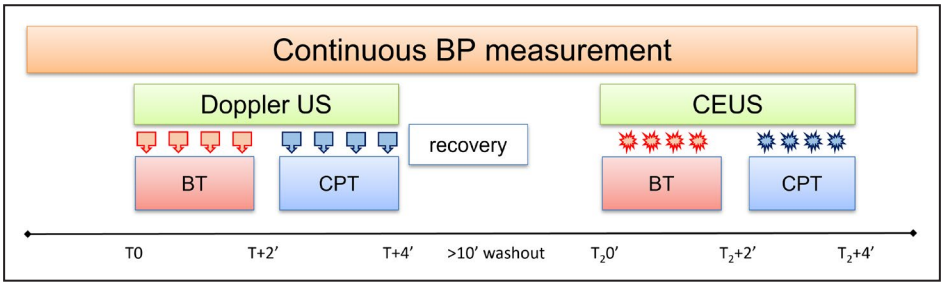

Figure 1. Study design.
Feet were placed in a water bath filled with water at BT for baseline Doppler US measurements. Thereafter, the water was replaced by cold water (2–4 °C) for 2 min for the measurements during the CPT. During the second phase, the same procedure was repeated for the CEUS measurements. BT indicates body temperature; CEUS, contrast-enhanced ultrasound; CPT, cold pressor test; and US, ultrasound.

expiratory breath-hold at baseline and during CPT with special care to select the same segmental artery at each measurement. RRI was calculated as (peak systolic velocity—end-diastolic velocity)/peak systolic velocity in the color Doppler ultrasound mode.

After a 10-minute recovery phase, a CEUS examination was performed at low mechanical index (0.08). The same kidney was chosen for image acquisition in a sagittal plane, and the probe was oriented so that renal length was highest, to guarantee the largest cortical surface area. The Sonovue contrast agent was injected as a continuous infusion with a special rotating pump (Vueject, Bracco SA, Milan, Italy) at a continuous rate of 0.015 mL/kg per min (≈1 mL/min for a 70-kg person). Image depth, focus, gain and frame rate were optimized and held constant during the experiment. Intermittent application of high-intensity sound waves to a kidney (destruction–replenishment technique) destroys the microbubbles locally; their reappearance rate in the cortex can be measured and expressed as the PI. As such, the PI is a quantitative measure of cortical microperfusion.[19] Once saturation was reached after 1 to 2 minutes, the destruction–replenishment technique was used to quantify intrarenal perfusion as described previously.[19,20] A minimum of 4 consecutive destruction–replenishment sequences were performed at each time point in breath-hold, to avoid movement artifacts.

Image Analysis

Images were exported in Digital Imaging and Communication in Medicine format and analyzed with the dedicated software Vuebox (Bracco Research, Geneva, Switzerland) using the replenishment model. Regions of interest were drawn manually and included the largest part of the renal cortex, as published previously.[19,21] Compensation of movement (minor breathing artifacts or small movements of the probe) was applied to all sequences before analysis. The Digital Imaging and Communication in Medicine data in each region of interest were converted into echo-power data that are directly proportional to the concentration of the contrast agent. For each region of interest, the rate at which microbubbles replenish renal tissue after destruction by pulses at high mechanical index is proportional to the local blood flow and is expressed as time–intensity curve. The mean transit time is the time needed, after microbubble destruction, to reach 50% of the maximal intensity signal (point at maximum slope or wash-in rate). The ratio of relative blood volume (rBV) to mean transit time allows the calculation of the PI (PI=rBV/mean transit time). Only sequences with a quality of fit of >85% between the echo-power signal and the time–intensity curve in a specific region of interest were used.

Statistical Analysis

Statistical analyses were performed using Stata software version 16 (StataCorp, College Station, TX). Data were summarized by group (women versus men) and by phase (baseline, CPT, recovery) and expressed as mean±SD for normally distributed data and as median and interquartile range for non-normally distributed data. We used a 2-sample *t* test or Wilcoxon rank-sum test or a χ^2 test to compare demographic data. Correlations between changes in renal and systemic hemodynamics during CPT were assessed by Spearman test. To evaluate the effect of the CPT in men and women on different hemodynamic and ultrasonographic outcomes, a linear mixed-effect model was used. Non-normally distributed outcomes were first log-transformed and analyzed on the logarithmic scale. Tested covariates include group (men versus women), phase (baseline, CPT, and recovery if measured) and their interaction. For sensitivity analysis, the same test was used in postmenopausal women only.

RESULTS

Participants

Thirty-two patients with hypertension, 11 women and 21 men, were included in the study. Age, body mass index, estimated glomerular filtration rate, and kidney length were similar in both groups. Baseline characteristics of the participants are presented in Table 1.

Continuous Systemic Hemodynamics

No difference was found in systolic BP, diastolic BP, and mean BP between men and women. The CPT increased BP in both groups as expected. During the recovery period, the systolic BP returned to baseline level, but diastolic BP and mean BP were slightly but significantly higher. There was no difference between men and women and no interaction between the stress test and sex. Age and body mass index had no

Table 1. Demographic Characteristics of the Participants by Sex

	Men (n=21)	Women (n=11)
Age, y	46.2±12.0	46.5±15.0
Currently smoking, n (%)	1 (4.8)	2 (18.2)
Body mass index, kg/m^2	26.8±4.2	25.4±3.4
Estimated glomerular filtration rate, mL/min per 1.73 m^2	94.6±19.8	93.9±19.5
Kidney length, mm	115±9	111±10
Kidney width, mm	48.6±6.8	43.5±5.2
Pulse wave velocity, m/s	7.49±1.22	7.47±1.62

Data are presented as mean±SD.

effect in the model. The CPT increased heart rate in both groups and returned to baseline value during the recovery period. No interaction was found between the CPT and the groups. Age was negatively associated with heart rate in univariate analysis only (β=−0.349; *P*=0.001). Age did not change the results of the mixed-model analysis. The results are shown in Table 2.

Hormones

At rest, norepinephrine, epinephrine, neuropeptide Y, plasma renin activity, and PAC levels were similar (Table S1). Norepinephrine and epinephrine levels increased after CPT in both groups as expected. No increase was found in plasma renin activity, PAC, and neuropeptide Y 1-36 after CPT. Age and body mass index had no effect on the model. Although there was no difference in PAC between the groups or with the CPT, the response to the CPT was different (significant interaction), with men increasing PAC as compared with women.

Doppler Ultrasound and CEUS Parameters

There was no difference in RRI, peak systolic velocity, or end-diastolic velocity between the groups or with the CPT. No interaction between the groups and the stress test was observed (Table 3).

Age was positively associated with RRI (β=0.004; *P*<0.001) and end-diastolic velocity (β=−0.097; *P*=0.025) in univariate analysis, but did not affect the results in the mixed model. The PI was higher in women (Figure 2) and increased during the CPT (Figure 3). When we corrected for body surface area, PI remained significantly higher in women. In a sensitivity analysis including only postmenopausal women (N=6), the PI was still higher in women but with borderline significance (*P*=0.079). When indexed to body surface area, the PI was higher in women (*P*=0.036). No interaction between the groups and the CPT was observed. Body mass index was negatively associated with PI in univariate analysis but did not affect the results in mixed analysis. No statistically significant correlations between BP changes and cortical PI were revealed for both men with hypertension (Spearman's rho=−0.22; *P*=0.34) and women with hypertension (Spearman's rho=0.09; *P*=0.80). The rBV was also higher in women and increased during CPT. No interaction was found between sex and the stress test. Finally, when we tested the relative increase (percentage) in cortical PI during CPT in women and men, no significant difference was found (50.61 [interquartile range, 28.8–81.1]) versus 25.90 ([interquartile range, −15.1 to 67.9]); (*P*=0.16).

DISCUSSION

The main finding of our study is that cortical PI reflecting microperfusion is higher in women with hypertension

Table 2. Changes in Systemic Hemodynamic During the CPT by Sex

		Baseline	CPT 120s	Recovery	Univariate analysis group, β (*P* value)	Univariate analysis phase β (*P* value)	Adjusted analysis, β (*P* value)	Adjusted analysis group×phase, β (*P* value)
Systolic BP, mm Hg	Men	150±15	164±17	154±18	−0.086 (1.0)	Baseline vs CPT 14.2 (<0.001) Baseline vs recovery 3.49 (0.2)	Group −0.358 (1.0) Baseline vs CPT 13.9 (<0.001) Baseline vs recovery 3.62 (0.2)	Baseline vs CPT No interaction Baseline vs recovery No interaction
	Women	150±17	164±17	153±15				
Diastolic BP, mm Hg	Men	93±13	104±16	97±15	−0.464 (0.9)	Baseline vs CPT 11.6 (<0.001) Baseline vs recovery 4.04 (<0.05)	Group −1.031 (0.8) Baseline vs CPT 10.96 (<0.001) Baseline vs recovery 4.11 (<0.05)	Baseline vs CPT No interaction Baseline vs recovery No interaction
	Women	92±10	105±8	96±10				
Mean arterial pressure, mm Hg	Men	115±14	128±17	120±16	1.14 (0.8)	Baseline vs CPT 13.1 (<0.001) Baseline vs recovery 4.19 (<0.05)	Group 0.77 (0.9) Baseline vs CPT 12.7 (<0.001) Baseline vs recovery 4.35 (0.1)	Baseline vs CPT No interaction Baseline vs recovery No interaction
	Women	116±12	130±10	120±11				
Heart rate, bpm	Men	61±8	66±12	62±9	3.26 (0.3)	Baseline vs CPT 5.86 (<0.001) Baseline vs recovery 0.373 (0.8)	Group 2.95 (0.4) Baseline vs CPT 4.74 (<0.001) Baseline vs recovery 1.233 (0.4)	Baseline vs CPT No interaction Baseline vs recovery No interaction
	Women	64±6	72±11	62±5				

Data are presented as mean±SD. BP indicates blood pressure; and CPT, cold pressor test.

Table 3. Changes in Doppler Ultrasound and CEUS Parameters During the CPT by Sex

		Baseline	CPT	Univariate analysis group, β (*P* value)	Univariate analysis phase, β (*P* value)	Adjusted analysis, β (*P* value)	Adjusted analysis group×phase, β (*P* value)
RRI, a.u.	Men	0.59±0.05	0.60±0.06	0. 027 (0.2)	0.005 (0.3)	Group 0.031 (0.2) Baseline vs CPT 0.0075 (0.2)	Baseline vs CPT No interaction
	Women	0.63±0.07	0.63±0.07				
Peak systolic velocity, cm/s	Men	31.0±8.7	31.4±7.9	4.97 (0.2)	0.728 (0.3)	Group 4.58 (0.2) Baseline vs CPT 0.453 (0.6)	Baseline vs CPT No interaction
	Women	35.5±15.0	36.8±13.7				
End-diastolic velocity, cm/s	Men	12.4±3.2	12.3±3.1	0.668 (0.6)	0.183 (0.5)	Group 0.324 (0.8) Baseline vs CPT −0.054 (0.9)	Baseline vs CPT No interaction
	Women	12.7±4.3	13.4±4.1				
PI, a.u.	Men	1285 (591–1741)	1813 (993–2330)	0.597 (<0.01)	0.350 (<0.001)	Group 0.674 (<0.01) Baseline vs CPT 0.403 (<0.001)	Baseline vs CPT No interaction
	Women	2344 (1553–2685)	2464 (2105–4134)				
PI corrected for body surface area, a.u.	Men	664 (308–797)	969 (474–1267)	0.751 (<0.01)	0.350 (<0.001)	Group 0.828 (<0.01) Baseline vs CPT 0.403 (<0.001)	Baseline vs CPT No interaction
	Women	1229 (875–1786)	1360 (1185–2463)				
rBV, a.u.	Men	2662 (2134–3983)	3459 (2173–5478)	0.415 (<0.05)	0.366 (<0.001)	Group 0.445 (<0.05) Baseline vs CPT 0.387 (<0.001)	Baseline vs CPT No interaction
	Women	3786 (2910–6452)	5620 (4442–6938)				
Mean transit time, s	Men	2.22 (2.05–3.27)	2.33 (1.96–3.07)	−0.146 (0.2)	0.0156 (0.8)	Group −0.208 (0.1) Baseline vs CPT −0.0268 (0.7)	Baseline vs CPT No interaction
	Women	1.56 (1.38–3.68)	2.30 (1.68–3.03)				

Data are presented as mean±SD or median (interquartile range). a.u. indicates arbitrary units; CPT, cold pressor test; PI, perfusion index; rBV, relative blood volume; and RRI, renal resistive index.

than in men with hypertension in resting-state conditions. When exposed to a CPT, both women and men with AH increase their renal cortical PI to a similar extent. This is the first report showing that women with hypertension have a higher PI than men with hypertension at rest. As the PI is dependent on rBV and mean transit time, this finding may be secondary to a higher rBV observed, which might reflect a more vasodilatory state.

We have shown previously that the PI in healthy participants is also higher in women.[14] Most studies have either included healthy men or included women and men but with suspected CKD.[19,22] An observational study including healthy controls, with a high proportion of women, and patients with diabetes across different stages of CKD, showed that the effect on sex was not significant on renal cortical perfusion assessed by arterial spin labeling magnetic resonance imaging.[23]

Renal plasma flow is ≈20% lower in women.[24] This difference observed with PI (higher in women) may be because renal plasma flow reflects the whole kidney perfusion (macrocirculation) and that cortical PI estimates cortical perfusion (microcirculation). CEUS-assessed perfusion seems to correlate with renal plasma flow in healthy (overweight or not) participants and in patients with various renal diseases; however, the associations were relatively weak, and the changes were not uniformly concordant[19,25,26] Finally, perfusion alterations can occur at the microcirculatory level without changes in large-vessel blood flow.

We can only hypothesize on the reasons why the cortical perfusion was higher in women with hypertension in our study. Sex-related differences in microvessel density of the renal cortex may account for the higher PI found in women. A rarefaction of microvessel density is closely associated with AH, metabolic syndrome, and type 2 diabetes.[27–29] A reduced bioavailability of vasodilators like NO has been hypothesized as an underlying mechanistic contributor. Interestingly, in models of AH, sex differences in microvessel density have been reported, with the male Sprague–Dawley rats exhibiting significant rarefaction at the gastrocnemius level when compared with female rats.[30] Moreover, a higher NO production has been reported in the systematic vasculature of women compared with men, which may partially account for the increased renal cortical perfusion.[31] Unfortunately, NO production was not measured in our study, so we cannot confirm or reject this

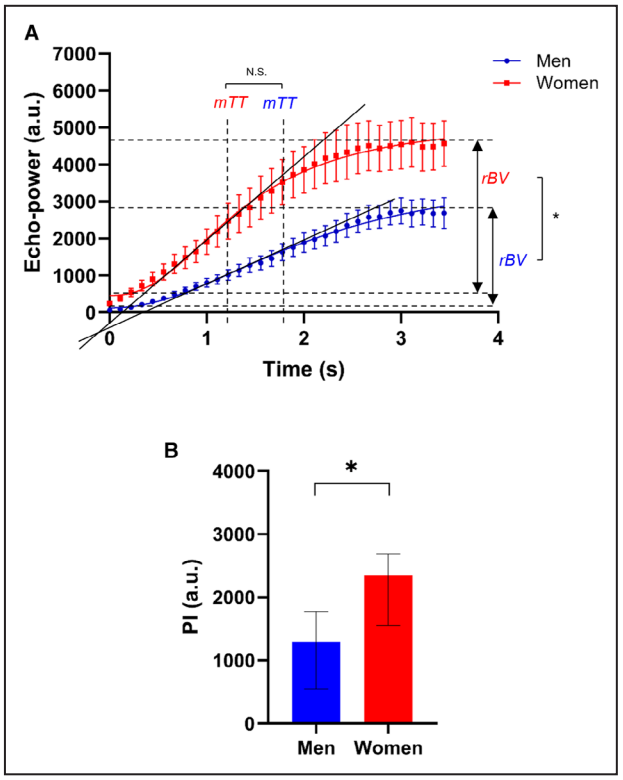

Figure 2. Cortical time intensity curve (A) and PI (B) in men and women at rest (baseline).
The mTT is the time from microbubbles destruction to 50% of the rBV; this corresponds to the point of the steepest slope of the time–intensity curve when the curve is sigmoid. The rBV corresponds to the maximum detected echo power (pixel luminescence). The (PI) is defined as rBV/mTT. PI data are presented as median (interquartile range). a.u. indicates arbitrary units; mTT, mean transit time; n.s., non-significant; PI, perfusion index; and rBV, relative blood volume; and s, seconds.

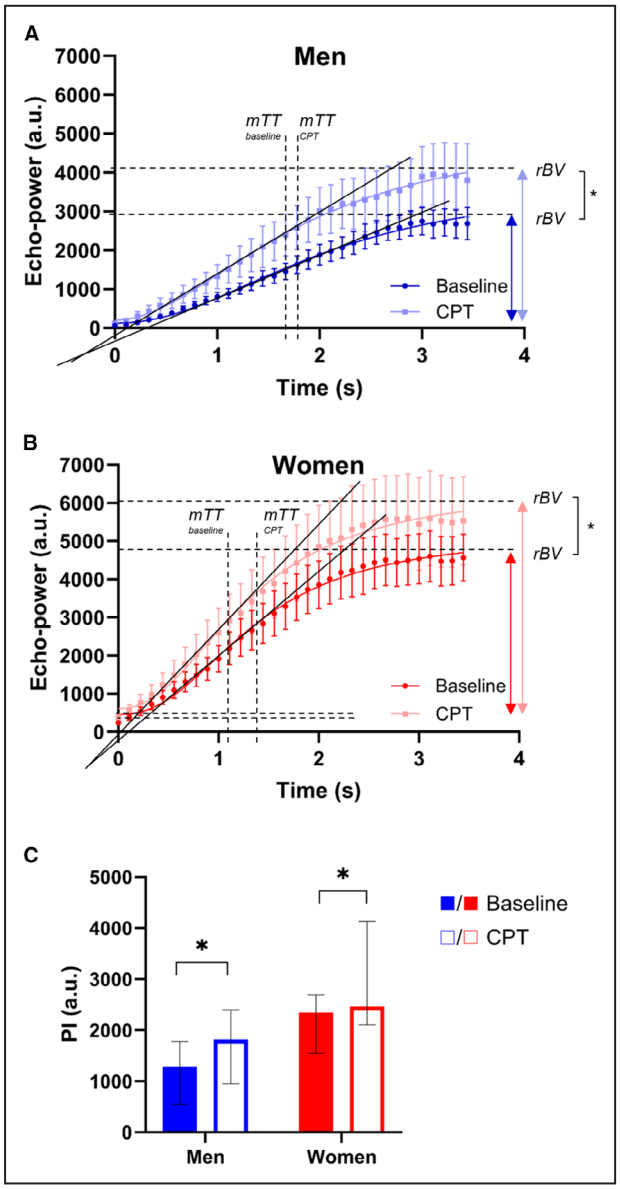

Figure 3. PI and its parameters at rest and during the experiment by sex.
Cortical time–intensity curve at rest and during CPT in men (**A**) and women (**B**). **C**, PI at rest and during CPT in men and women. PI data are presented as median (interquartile range). a.u. indicates arbitrary units; CPT, cold pressor test; PI, perfusion index.

hypothesis. At the kidney level, a recent animal study using histopathological examination demonstrated increased microvascular density in female mice and variations in the expression of estrogen receptor β across different strains, emphasizing the roles of both sex and genetic factors.[32] Future studies with larger participant cohorts are needed to determine whether these sex-specific differences in the microcirculation impact the prevalence, progression, and outcomes of conditions such as hypertension, CKD, or acute kidney injury.

Several epidemiologic studies have highlighted that premenopausal women are preserved from AH and end-stage kidney disease. Protection against these disease states has been attributed partially to estrogen, which exhibits protective effects on the endothelium, while androgens may be antagonistic to cardiovascular health. Additionally, there is evidence that estrogens exert antioxidative and antifibrotic effects in CKD models and that they stimulate renal vasodilation by enhancing NO production by glomerular cells.[31,33,34] In line with this, postmenopausal women display cardiovascular risk profiles that are similar to age-matched men.[35] However, other inherent cardiovascular advantages might exist, since a study showed that in ovariectomized animal models, women were still protected against renal vasoconstriction and glomerular sclerosis.[36] In our study the effect of sex hormones on PI is only speculative as almost half of the women (6/11) who participated in our study were postmenopausal. When we removed the premenopausal women in the mixed-model analysis (Table S2), baseline PI was still higher in women when indexed to body surface

area, and just above significance level when not indexed. The low number of postmenopausal women (N=6) may have decreased the power of this analysis. In the aforementioned study including healthy participants, the phase of the menstrual cycle did not alter cortical perfusion.[14] Finally, it has been shown that the renal vasculature of men becomes more dependent on NO, especially with age, compared with women.[37,38] Nevertheless, the higher PI remained significant in women with hypertension in the age-matched analysis.

Sex differences have been also reported in the endothelin system regarding the endothelin receptors' (endothelin A and endothelin B) expression and activation, the levels of endothelin-1, a potent vasoconstrictor, and the downstream mediators.[39] Interestingly, in a swine model of renovascular disease, endothelin A receptor blockade improved renal perfusion and reversed microvascular rarefaction as well as when the blockade was combined with percutaneous transluminal renal angioplasty.[40,41]

On the other hand, sex hormones have been implicated for the different levels of endothelin-1 and the different sensitivity to endothelin-1 vasoconstriction found between sexes in animal and human studies.[39] Men seem to exhibit greater endothelin A–mediated vasoconstriction than women, whereas women exhibit enhanced endothelin B–mediated vasodilation. In addition, there is evidence of sex differences in the contribution of endothelin B receptor activation to modulate renal function.[42] Taken together, this sexual dimorphism in the endothelin system could mediate a higher vascular tone in the resting state and thus could partially explain the lower PI found in men with hypertension compared with women with hypertension. Additionally, a greater endothelin-1/endothelin A receptor activation in men may result in increased oxidative stress and thus contribute to a more burdened and less preserved renal and microvasculature function.

The increase in renal cortical perfusion during CPT was similar in both sexes. This increase suggests vasodilation rather than vasoconstriction as we have previously reported.[11] This finding underlines that the capacity to augment flow upon stimulation does not seem to be sex specific, with both women with hypertension and men with hypertension reacting to the same degree to a sympatho-excitatory stressor. This is supported by a previous animal study showing that there were no differences in the responsiveness to endothelium-dependent or -independent vasodilators between the male and female rat kidneys.[43] Indeed, the similarity of the vasodilator response to acetylcholine between male and female rats suggest that sex does not have an impact on endothelial responsiveness to vasodilators in the resistance arteries of the rat kidney. In addition, even in a setting where functional rarefaction might be present, the non-perfused capillaries at rest can be recruited and still have the potential to participate in perfusion when responding to increased metabolic demands.[44] Finally, the response to a sympatho-excitatory stress does not seem to follow the sex difference observed when aldosterone response to angiotensin II perfusion is compared in men and women.[45]

In contrast with our findings, Kannenkeril et al reported a reduction of cortical renal blood flow measured by arterial spin labeling magnetic resonance imaging with the same stress test (CPT) in both patients with hypertension and patients without hypertension.[46] However, in this study, no women were included.

To the best of our knowledge, an increase in cortical PI has not been reported as harmful or associated with disease states that are characterized by hyperfiltration such as diabetes, obesity, or hypertension. On the other hand, in settings such as CKD and acute kidney injury, a decrease in cortical microperfusion has been consistently reported.[10,47,48]

The limitations of our study include the absence of randomization of the sequences regarding CPT exposure, which may have induced tolerance to the cold exposure. However, the magnitude of BP increase with each stimulation was similar, which speaks against such an effect. Another limitation was the small sample size as well as the higher proportion of men who participated in the study. Furthermore, while the observed differences in PI remained robust even after adjusting for kidney size, the potential for type I error cannot be completely excluded given the small sample size and the multiple comparisons made. Finally, the absence of hormonal data limits our ability to conclude on the effect of sex hormones on the cortical microperfusion and to fully account for other potential confounders that could influence the PI.

In conclusion, our study demonstrates the presence of sex differences in the cortical microcirculation as assessed by CEUS, with women with hypertension exhibiting a higher PI at resting state. Finally, both sexes react to the same extent to CPT, pointing out that there is no sex-specific difference in reactivity of the renal microvasculature to sympatho-excitatory stressors. Future studies implementing bedside techniques such as CEUS will promote a more comprehensive anatomic and functional picture of the female and male renal microvasculature in disease states in which the relevance of renal hemodynamics is of high importance.

ARTICLE INFORMATION

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Disclosures

None.

Supplemental Material

Tables S1–S2

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