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Chronic hydrogen-rich saline treatment reduces oxidative stress and attenuates left ventricular hypertrophy in spontaneous hypertensive rats

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Abstract In hypertensive animals and patients, oxidative stress represents the primary risk factor for progression of left ventricular hypertrophy. Recently, it has been demonstrated that hydrogen, as a novel antioxidant, can selectively reduce hydroxyl radicals and peroxynitrite anion to exert therapeutic antioxidant activity. In the current study, we explored the effect of chronic treatment with hydrogen-rich saline (HRS) on left ventricular hypertrophy in spontaneously hypertensive rats (SHR). The 8-week-old male SHR and age-matched Wistar-Kyoto rats (WKY) were randomized into HRS-treated (6 ml/kg/day for 3 months, i.p.) and vehicle-treated groups. HRS treatment had no significant effect on blood pressure, but it effectively attenuated left ventricular hypertrophy in SHR. HRS treatment abated oxidative stress, restored the activity of antioxidant enzymes including GPx, GST, catalase, and SOD, suppressed NADPH oxidase activity and downregulated Nox2 and Nox4 expression in left ventricles of SHR. HRS treatment suppressed pro-inflammatory cytokines including IL-1β, IL-6, TNF-α, and MCP-1, and inhibited NF-κB activation through preventing IκBα degradation in left ventricles of SHR. HRS treatment preserved mitochondrial function through restoring electron transport chain enzyme activity, repressing ROS formation, and enhancing ATP production in left ventricles of SHR. Moreover, HRS treatment suppressed ACE expression and locally reduced angiotensin II generation in left ventricles of SHR. In conclusion, HRS treatment attenuates left ventricular hypertrophy through abating oxidative stress, suppressing inflammatory process, preserving mitochondrial function, in which suppression of HRS on angiotensin II in left ventricles locally might be involved.

Keywords Hydrogen-rich saline · Spontaneously hypertensive rats · Oxidative stress · Inflammation · Mitochondria · Angiotensin II

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Introduction

Hypertensive patients and animals exhibit left ventricular hypertrophy [1, 2], which is a compensatory response and often results in poor clinical outcomes, including the development of cardiac systolic and diastolic dysfunction and ultimately heart failure [3–5]. Growing evidence implicates that oxidative stress produced by overproduction of reactive oxygen species/reactive nitrogen species or inefficient antioxidant defenses appears to be involved in the development of hypertension-induced left ventricular hypertrophy [6, 7]. However, clinical trials with antioxidant vitamin C and E failed to show an improved cardiovascular outcome and larger prospective randomized trials in humans looking at hard cardiovascular end-points have been disappointing [8, 9]. Does this mean that antioxidant approaches have no role in prevention or treatment in cardiovascular diseases?. In fact, it is clear that the term oxidative stress covers a diverse array of complex biological actions ranging from highly specific redox signaling involving specialized enzymes to interactions with nitric oxide to more straightforward effects such as ''damage'' to cells, membranes and

macromolecules. Therefore, it is hardly surprising that the use of non-specific antioxidants did not produce the desired results. Moreover, these agents are relatively inefficient and have generally been used without any assessment of the target population for levels of oxidative stress. What may be required are much more specific and more efficient agents that can target defined ROS sources and deleterious redox-dependent signal pathways.

Recently, it has been proved that hydrogen gas, a highly flammable gas, has potent antioxidant property. Compared with normal antioxidants such as vitamin C and vitamin E, hydrogen has two unique characteristics. First, it selectively reduces the hydroxyl radical (−OH) and peroxynitrite anion ($ONOO^-$), the most cytotoxic chemicals of ROS, and effectively protects cells; in addition, hydrogen dose not react with other ROS, which possess physiological roles [10]. Second, since the hydrogen molecule is electrically neutral and much smaller than the other antioxidants, it is able to easily penetrate membranes and enter cells and organelles, such as the nucleus and mitochondria, where most commonly used antioxidants cannot arrive [11].

It has been demonstrated that treatment with hydrogen gas or hydrogen-rich saline (HRS) protects organs damage such as transient cerebral ischemia [12], neonatal cerebral hypoxia–ischemia [13], renal injury [14] and myocardial injury induced by ischemia and reperfusion [15]. However, most of these researches focus on the effect of short-term treatment with hydrogen on acute injury. In the present study, we explored whether chronic treatment with HRS attenuated left ventricular hypertrophy in spontaneously hypertensive rats.

Methods

Animals and study design

The 8-week-old male SHR and age-matched normotensive Wistar-Kyoto rats (WKY) were obtained from the Sino-British SIPPR/BK Lab Animal Ltd. All the animals were entrained to controlled temperature (23–25°C), 12-h light and 12-h dark cycles (light 08:00–20:00 h; darkness 20:00–08:00 h), and free access to food and tap water. All the animals used in this study received humane care in compliance with institutional animal care guidelines, and were approved by the Local Institutional Committee. All the surgical and experimental procedures were in accordance with institutional animal care guidelines. Chemicals, reagents, and drugs were purchased from Sigma Chemical (St. Louis, MO, USA) unless otherwise stated.

Animals were divided into four groups and treated for 3 months as follows: (1) WKY rats treated with saline; (2) WKY rats treated with the HRS; (3) SHR treated with saline; (4) SHR treated with the HRS. 6 ml/kg of HRS or saline was intraperitoneally (i.p.) injected daily in the morning.

Hydrogen-rich saline (HRS)

Two methods including inhalation of hydrogen gas or injection of HRS were usually utilized for treatment. Compared to hydrogen gas, HRS is safe, economical, easily available, and more convenient for long-term treatment. In current study, hydrogen was dissolved in physiological saline for 4 h under the pressure of 0.4 MPa as method described by Ohsawa et al. [10]. HRS was freshly prepared every week to insure a constant concentration more than 0.6 mM.

Blood pressure measurement

24 h after last treatment, systolic blood pressure (SBP), diastolic blood pressure (DBP), and heart rate (HR) were recorded continuously as previously described [16]. In brief, rats were anesthetized with a combination of ketamine (40 mg/kg, i.p.) and diazepam (6 mg/kg, i.p.). A floating polyethylene catheter was inserted into the lower abdominal aorta via the left femoral artery for BP measurement, and another catheter was indwelled in the left femoral vein for intravenous injection. The catheters were exteriorized through the interscapular skin. After a 2-day-recovery period, the animals were placed for BP recording in individual cylindrical cages with food and water. The aortic catheter was connected to a BP transducer via a rotating swivel that allows the animals to move freely in the cage. After about 14-h habituation, the BP signals were digitized by a microcomputer, and beat-to-beat SBP, DBP, and HR values were determined on line. The mean values of these parameters during the 24 h were calculated and served as SBP, DBP, and HR for study.

Sample preparation

The animals were anesthetized and killed by decapitation. Immediately after decapitation, a 2-ml blood sample was collected and heart was immediately excised and rinsed in cold physiological saline.

Assessment of left ventricular hypertrophy

Body and left ventricular weights were recorded. Transversely sectioned left ventricle frozen tissue sections (6 μm) were stained with Alexa Fluor® 594 wheat germ agglutinin and blue-fluorescent Hoechst 33342 dye (Invitrogen, Carlsbad, CA, USA). Four radially oriented

microscopic fields from each section were photographed and the cross-sectional area of at least 100 cells, in which the nucleus and a clear staining of the plasma membrane could be visualized, were averaged. The myocyte outlines were traced, and the cell areas were measured using "lasso" tool in Adobe Photoshop.

Measurement of left ventricular and serum malondialdehyde (MDA)

MDA concentration is a presumptive marker of oxidant-mediated lipid peroxidation. Left ventricular homogenates and serum were used for the determination of MDA using a kit (Cayman, Ann Arbor, USA).

Western blotting analysis

The protein concentration was determined with bovine serum albumin as a standard by a Bradford assay. Equal amounts of protein preparations (10 μg in 10 μl buffer) were run on SDS-polyacrylamide gels, electrotransferred to polyvinylidine difluoride membranes, and blotted with a primary antibody against Nox2 (1:1000, Abcam, Cambridge, UK), Nox4 (1:500, Abcam), ACE (1:500, Abcam) overnight at 4°C using slow rocking. Then, they were blotted with HRP-conjugated secondary antibody (1:5000) and HRP-conjugated monoclonal antibody against GAPDH (1:10000). Immunoreactive bands were detected by a chemiluminescent reaction (ECL kit, Amersham Pharmacia), and results were expressed as the ratio of the density of specific bands to the corresponding GAPDH. The WKY group was used as the calibrator with a given value of 100%, and the other groups were compared with this calibrator.

NADPH oxidase activity

Lucigenin-enhanced chemiluminescence was used to measure NADPH oxidase activity in left ventricles according to the method described previously [17].

Determination of SOD activity, catalase activity, glutathione-S-epoxide transferase (GST) activity, and glutathione peroxidase (GPx) activity in left ventricles

SOD activity was measured using an SOD-525TM reagent kit (OXIS International, Foster, CA, USA). Catalase activity was measured by the method of Beers and Sizer, as previously described [18]. GST activity was measured using GST Fluorometric Activity Assay Kit (BioVision, Mountain View, CA, USA). GPx activity was determined in left ventricular homogenates using a commercially available kit, according to the manufacturer's protocol (Cayman Chemicals, Ann Arbor, MI, USA). The final results were corrected for protein content.

Total ROS and $OONO^-$ production in left ventricles

Total ROS and $OONO^-$ productions were detected as per the method described by Elks et al. [19].

Measurement of mitochondrial ATP production and ROS formation

Mitochondria were isolated by differential centrifugation of left ventricular homogenates. Mitochondrial protein concentration was determined using a DC Protein Assay Kit (Bio-Rad, Hercules, CA, USA). Rates of ATP formation were quantified using a commercially available kit (BioVision, Mountain View, CA, USA). Mitochondrial ROS production was evaluated by lucigenin chemiluminescence. The results were corrected for protein content.

Measurement of mitochondrial electron transport chain enzyme activity

The activity of rotenone-sensitive complex I, malonate-sensitive complex II, antimycin A-sensitive complex III, and sodium cyanide-sensitive complex IV, as well as reduced nicotinamide-adenine dinucleotide cytochrome c reductase (NCCR; marker for electron coupling capacity between complexes I and III) or succinate cytochrome c reductase (SCCR; marker for electron coupling capacity between complexes II and III), were assayed [20] using a thermostatically regulated Thermo-Spectronic spectrophotometer (Fisher Scientific, CA, USA).

ANG II measurement in plasma and left ventricles

Plasma and left ventricular samples were used for the measurement of ANGII using radioimmunoassay kits (China Institute of Atomic Energy, Beijing, China).

Quantitative real-time PCR analysis (RT-PCR)

Total RNA was extracted from left ventricles using TRIzol (Life Technologies Inc., Gaithersburg, USA) according to the manufacturer's protocol. Real-time PCR analysis was performed with a QuantiTectTM SYBR® Green PCR (Tiangen, Shanghai, China) according to the manufacturer's instructions. The sequences of primers are listed in Table 1. The highly specific measurement of mRNA was carried out for TNF-α, IL-6, IL-1β, IκBα, NF-κB p65 and GAPDH using the LightCycler system (Bio-Rad, Carlsbad, USA). TNF-α, IL-6, IL-1β, IκBα, and NF-κB p65 mRNA levels were adjusted as the values relative to GAPDH,

Table 1 Sequences of oligonucleotides used as primers

Target gene		Sequence (5′–3′)	Accession number[a]
IL-6	Sense	TCCTACCCCAACTTCCAATGCTC	NM_012589
	Antisense	TTGGATGGTCTTGGTCCTTAGCC	
IL-1β	Sense	CACCTCTCAAGCAGAGCACAG	NM_031512
	Antisense	GGGTTCCATGGTGAAGTCAAC	
TNF-α	Sense	CCAGGAGAAAGTCAGCCTCCT	X66539
	Antisense	TCATACCAGGGCTTGAGCTCA	
IκBα	Sense	CCCTGGAAAATCTTCAGACG	NM_001105720
	Antisense	ACAAGTCCACGTTCCTTTGG	
NF-κB p65	Sense	CATCAAGATCAATGGCTACA	NM_199267.2
	Antisense	CACAAGTTCATGTGGATGAG	
GAPDH	Sense	AGACAGCCGCATCTTCTTGT	NM_017008
	Antisense	CTTGCCGTGGGTAGAGTCAT	

[a] Genbank accession number of cDNA and corresponding gene is available at http://www.ncbi.nlm.nih.gov/

which was used as the endogenous control to insure equal starting amounts of cDNA. The WKY untreated by HRS was used as the calibrator with a given value of 1, and the other groups were compared with this calibrator.

Measurement of MCP-1 in left ventricles

The contents of MCP-1 in left ventricular homogenates with equal amounts of total protein were determined using a specific sandwich enzyme immunometric assay kit for rat MCP-1 (Assay Design, Ann Arbor, MI). All the samples were assayed in duplicate according to the manufacturer's instructions. The final result was corrected for protein content.

Statistical analysis

All data are presented as mean ± standard deviations. Comparison between groups was analyzed by two-way ANOVA. A probability level of less than 0.05 was considered significant.

Results

Hemodynamic parameters

SHR exhibited higher SBP, DBP, and HR compared with WKY (Table 2). Treatment with HRS had no significant effect on DBP and HR in both strains. Treatment with HRS decreased SBP in SHR, but not significantly.

Left ventricular hypertrophy

LVW-to-BW ratio in SHR was higher than it in WKY, indicating that SHR developed left ventricular hypertrophy (Fig. 1a). Average cardiomyocyte area of left ventricles of SHR was larger than that of WKY, revealing that cardiomyocyte hypertrophied in SHR (Fig. 1b). Treatment with HRS reduced LVW-to-BW ratio and cardiomyocyte area, indicating that HRS treatment attenuated left ventricular hypertrophy in SHR.

Oxidative stress

Compared with WKY, serum MDA content (Fig. 2a), and left ventricular MDA content (Fig. 2b), ROS (Fig. 2c) and $OONO^-$ (Fig. 2d) formation were higher in SHR, which were reversed after treatment with HRS.

Antioxidant enzymes

Compared with WKY, left ventricular GPx (Fig. 3a), GST (Fig. 3b), catalase (Fig. 3c), and SOD (Fig. 3d) activities were lower in SHR. Treatment with HRS restored them, at least in part.

Table 2 Effects of HRS on blood pressure and heart rate in WKY and SHR

	WKY	WKY + HRS	SHR	SHR + HRS
SBP (mmHg)	141 ± 16	143 ± 22	184 ± 19*	170 ± 15*
DBP (mmHg)	94 ± 14	92 ± 12	115 ± 16*	112 ± 10*
HR (bpm)	345 ± 23	340 ± 26	395 ± 29*	390 ± 25*

Values are represented as mean ± SD. *SBP* systolic blood pressure; *DBP* diastolic blood pressure; *HR* heart rate; *HRS* hydrogen-rich saline

* $P < 0.05$ versus WKY

$n = 11$ in each group

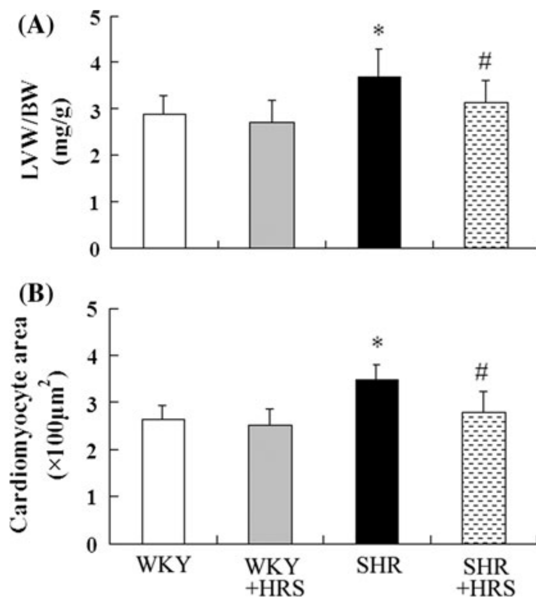

Fig. 1 Effect of HRS on left ventricular hypertrophy. Column graphs show LVW-to-BW ratio (**a**) and cardiomyocyte area (**b**) in left ventricles. $n = 11$ in each group. *LVW* left ventricular weight; *BW* body weight; *HRS* hydrogen-rich saline; Values are means ± SD. *$P < 0.05$ versus WKY; $^{\#}$ $P < 0.05$ versus SHR

NADPH oxidase

Compared with WKY, left ventricular NADPH oxidase activity (Fig. 4a) was increased in SHR. Furthermore, the Nox2 and Nox4 expression levels (Fig. 4b, c) in left ventricles in SHR were higher than in WKY. Treatment with HRS in SHR suppressed NADPH oxidase activity and decreased Nox2 expression, but had no significant effect on Nox4 expression.

Mitochondrial function in left ventricles

Compared to WKY, activities of complex I (Fig. 5a) and III (Fig. 5c), but not complex II (Fig. 5b) or IV (Fig. 5d) were significantly lower in left ventricles of SHR. There was also a significant decline in the electron-coupling capacity between complexes I and III (Fig. 5e) or between complexes II and III (Fig. 5f) in SHR, as demonstrated by the reduced activity of NCCR (Fig. 5g) or SCCR (Fig. 5h). In addition, mitochondria in left ventricles of SHR exhibited higher ROS formation and lower ATP formation. Treatment with HRS restored activities of complex I and III and electron-coupling capacity between complexes I and III and between complexes II and III, suppressed ROS formation, and increased ATP formation.

Pro-inflammatory cytokines in left ventricles

Compared with WKY, IL-1β (Fig. 6a), IL-6 (Fig. 6b), TNF-α (Fig. 6c) mRNA levels, and MCP-1 (Fig. 6d) content in left ventricles were higher in SHR, which were reversed after treatment with HRS.

NF-κB pathway

Compared with WKY, IκBα (Fig. 7a) mRNA level was decreased, while NF-κB p65 (Fig. 7b) mRNA level was increased in left ventricles of SHR. Furthermore, NF-κB p65 activity (Fig. 7c) in left ventricles of SHR was higher than that of WKY. Treatment with HRS restored them in left ventricles of SHR.

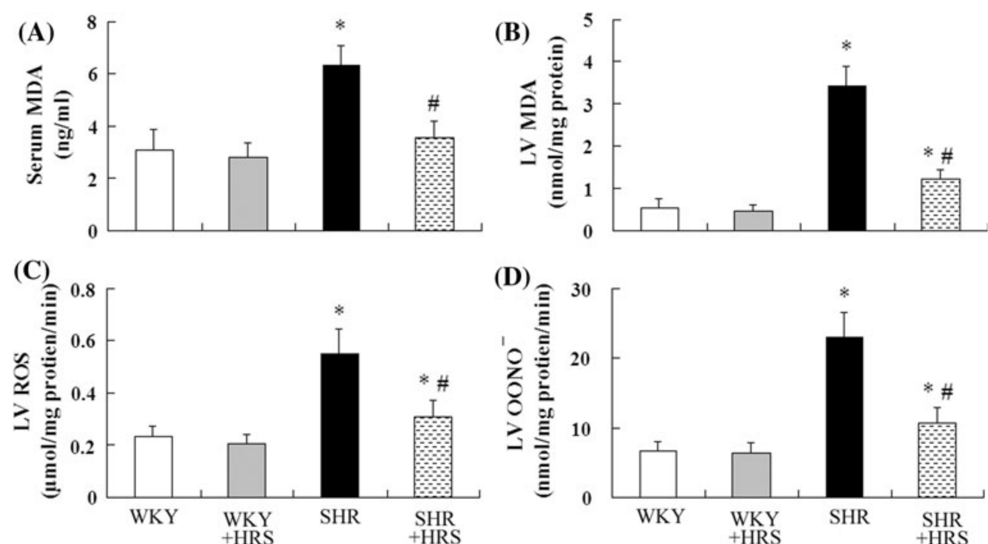

Fig. 2 Effect of HRS on oxidative stress. Column graphs show serum MDA (**a**), left ventricular MDA (**b**), ROS formation (**c**), and $OONO^{-}$ formation (**d**). $n = 11$ in each group. *LV* left ventricular; *MDA* malondialdehyde; *ROS* reactive oxygen species; *$OONO^{-}$* peroxynitrite; *HRS* hydrogen-rich saline; Values are means ± SD. *$P < 0.05$ versus WKY; $^{\#}$ $P < 0.05$ versus SHR

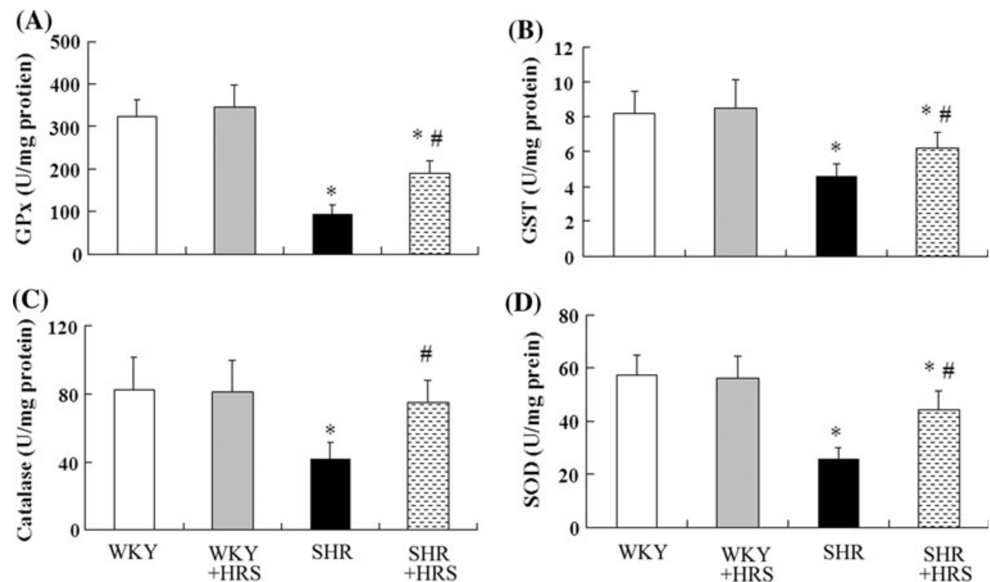

Fig. 3 Effect of HRS on antioxidant enzymes in left ventricles. Column graphs show GPx (**a**), GST (**b**), catalase (**c**), and SOD (**d**) activities in left ventricles. $n = 11$ in each group. *SOD* superoxide dismutase; *GST* glutathione S epoxide transferase; *GPx* glutathione peroxidase; *HRS* hydrogen-rich saline; Values are means ± SD. $*P < 0.05$ versus WKY; $^{\#}$ $P < 0.05$ versus SHR

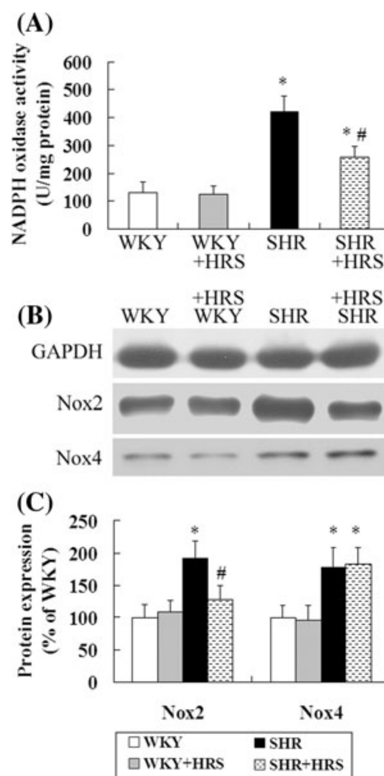

Fig. 4 Effects of HRS on NADPH oxidase activity and expression in left ventricles. Lucigenin-enhanced chemiluminescence result on enzyme activity of NADPH oxidase (**a**). $n = 11$ in each group. Western blot results (**b**) and responding quantification (**c**) of Nox2 and Nox4. The WKY group was used as the calibrator with a given value of 100%, and the other groups were compared with this calibrator. *HRS* hydrogen-rich saline; Values are means ± SD. $*P < 0.05$ versus WKY; $^{\#}$ $P < 0.05$ versus SHR

Angiotensin II (Ang II)

Compared with WKY, Ang II levels of plasma (Fig. 8a) and left ventricles (Fig. 8b) were higher in SHR. Furthermore, left ventricular ACE expression (Fig. 8c, d) was higher in SHR than in WKY. Treatment with HRS decreased ACE expression and Ang II level in left ventricles, but had no significant effect on Ang II level of plasma.

Discussion

In the current study, we examined the effects of long-term treatment with HRS on left ventricular hypertrophy in SHR. The salient findings of present study were summarized as follows: (1) Treatment with HRS alleviated left ventricular hypertrophy in SHR without a reduction of blood pressure level. (2) Treatment with HRS abated oxidative stress through upregulating activities of anti-oxidant enzymes and suppressing NADPH oxidase activity and mitochondrial ROS formation. (4) Treatment with HRS suppressed inflammation through decreasing pro-inflammatory molecules including IL-6, TNF-α, IL-1β, and MCP-1 in left ventricles of SHR. (5) Treatment with HRS exerted inhibitory effects on NF-κB by impeding IκBα degradation in left ventricles of SHR. (6) Treatment with HRS decreased left ventricular Ang II formation through suppressing ACE expression in SHR, but had no significant effect on circulating Ang II.

Oxidative stress plays an important role in the pathogenesis of left ventricular hypertrophy [21, 22]. In the present study, it was found that hydrogen not only neutralized with the toxic ROS marked by decreased $OONO^{-}$, but also influenced the antioxidant systems and pro-oxidant enzymes. It restored the activities of GST, GPx, SOD, and catalase, and suppressed the ROS formation derived from NADPH oxidase in left ventricles of SHR. In addition, HRS treatment exerted beneficial effect on mitochondrial

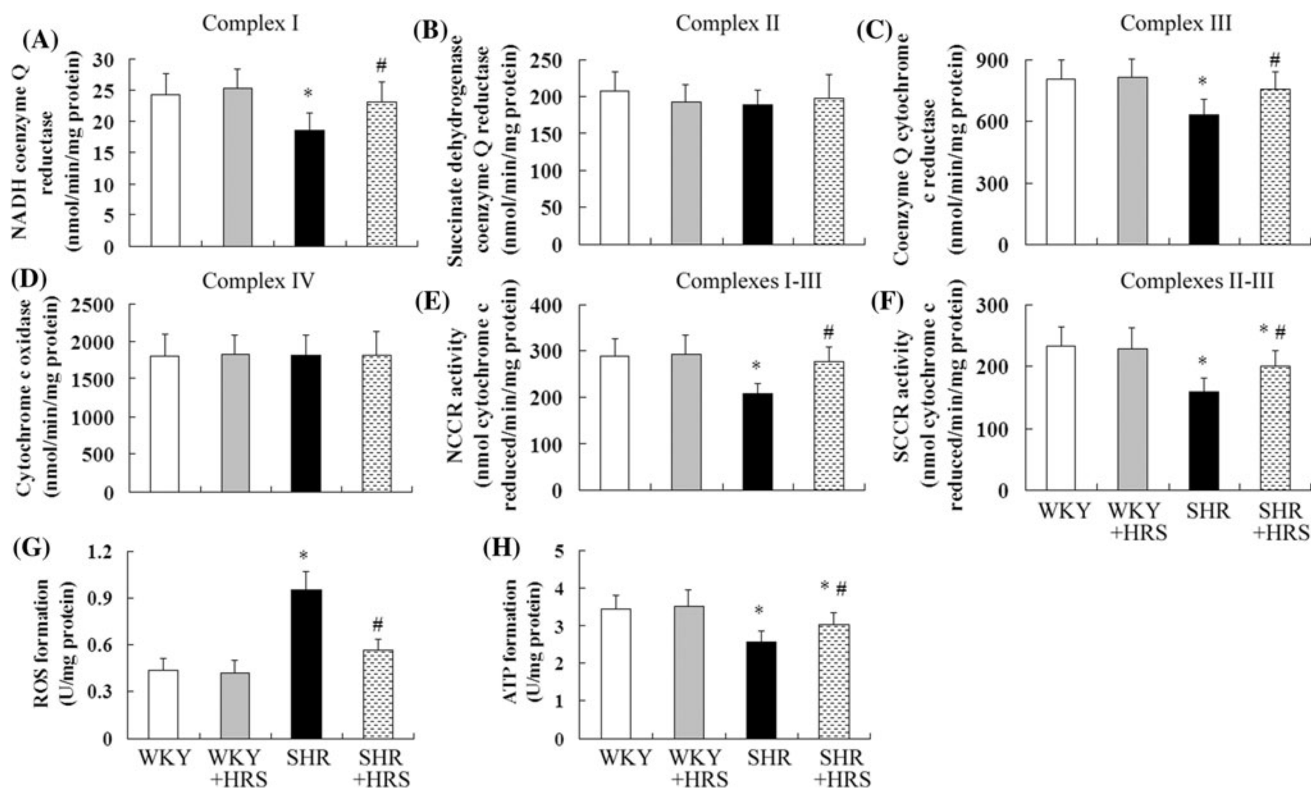

Fig. 5 Effect of HRS on mitochondrial function in left ventricles. Enzyme activities of complexes I to IV of mitochondrial electron transport chain (**a–d**) or electron coupling capacity between complexes I and II or complexes II and III, as denoted by the activity of NCCR or SCCR (**e**, **f**) and mitochondrial ROS formation (**g**) and ATP formation (**h**) in left ventricles. Values are means ± SD. $n = 11$ in each group. *HRS* hydrogen-rich saline; *SCCR* ; *NCCR* nicotinamide-adenine dinucleotide cytochrome c reductase; $^{*}P < 0.05$ versus WKY; $^{\#}$ $P < 0.05$ versus SHR

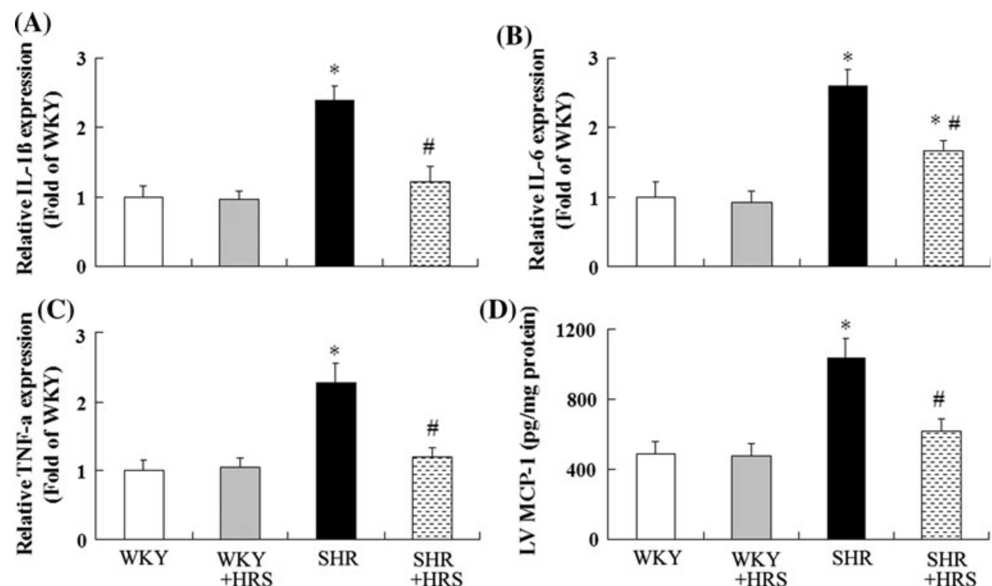

Fig. 6 Effect of HRS on pro-inflammatory cytokines in left ventricles. RT-PCR results on IL-1β (**a**), IL-6 (**b**), and TNF-α (**c**) mRNA expression, and MCP-1 concentration (**d**) in left ventricles. Values are means ± SD. $n = 11$ in each group. *HRS* hydrogen-rich saline; *LV* left ventricular; *MCP-1* macrophage chemoattractant protein 1; $^{*}P < 0.05$ versus WKY; $^{\#}$ $P < 0.05$ versus SHR

function. Recently, increasing evidence linked mitochondrial dysfunction and mitochondria-derived ROS to the pathogenesis of cardiovascular diseases [23–25]. Here, we found that HRS treatment restored mitochondrial electron transport chain enzyme activity, suppressed mitochondrial ROS formation, and enhanced ATP formation in left ventricles of SHR. The protective effect of HRS on mitochondria might be due to its effect on NADPH oxidase.

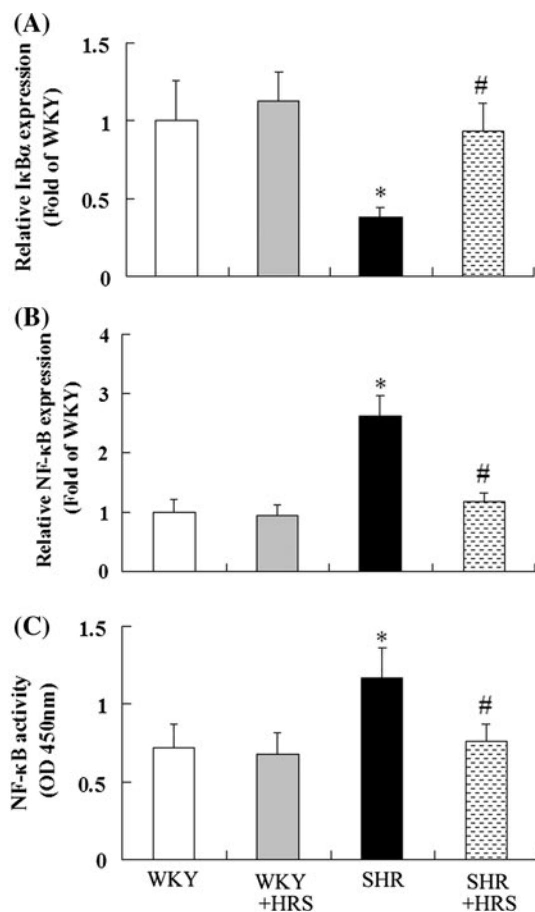

Fig. 7 Effect of HRS on NF-κB pathway in left ventricles. RT-PCR results on IκBα (**a**) and NF-κB p65 (**b**) mRNA expression in left ventricles and NF-κB activity (**c**) in nuclear extract of the left ventricles. *HRS* hydrogen-rich saline; *RT-PCR* reverse transcription polymerase chain reaction; *NF-κB* nuclear factor-kappa B; Values are means ± SD. $n = 11$ in each group. *$P < 0.05$ versus WKY; # $P < 0.05$ versus SHR

Several studies have revealed that NADPH oxidase-derived ROS lie upstream of mitochondria-produced ROS, and after mitochondrial dysfunction [26, 27]. However, since the hydrogen molecule is electrically neutral and much smaller than the other antioxidants, it is able to easily penetrate membranes and enter mitochondria to neutralize the excessive ROS. The unique property of hydrogen might also contribute to its protective effect on mitochondria.

Apart from antioxidant property, hydrogen exerted anti-inflammatory property in this study, which was also observed in several previous studies [28, 29]. Inflammation also plays a pivotal role in the progression of hypertensive left ventricular hypertrophy [30]. Here, we found that treatment with HRS suppressed pro-inflammatory cytokines including IL-6, TNF-α, IL-1β, and MCP-1 in left ventricles of SHR. The anti-inflammatory effect of hydrogen might be secondary to its anti-oxidative effect. Excess ROS activate the redox-sensitive transcription factor NF-κB, resulting in enhancement of its expression and activity [31, 32]. Increased expression and activity of NF-κB induces gene transcription of pro-inflammatory cytokines, such as IL-6 and IL-1β, to increase their production [31, 33]. In this study, left ventricular NF-κB activation in SHR was inhibited by treatment with HRS, which might interpret the anti-inflammatory property of HRS, at least in part.

In the current study, it was first observed that HRS treatment in SHR locally suppressed Ang II generation in left ventricles of SHR. Many reports have demonstrated important roles of Ang II in the cardiovascular system under pathological conditions, such as hypertension and heart failure [34, 35]. Ang II was implicated in the development of cardiomyocyte hypertrophy in humans as well

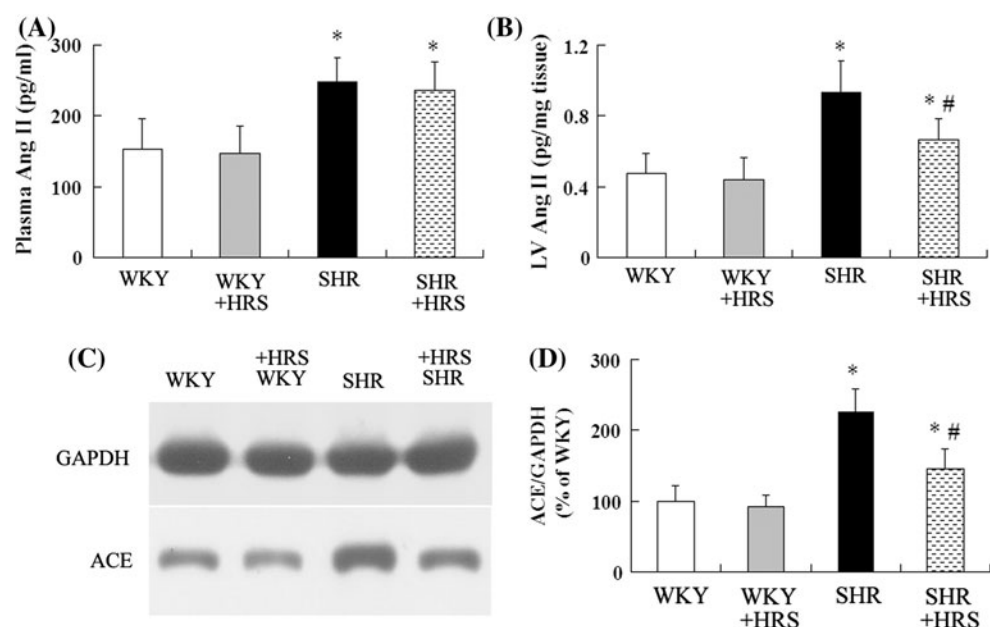

Fig. 8 Effects of HRS on Ang II formation and ACE expression. Radioimmunoassay results on plasma (**a**) and left ventricular (**b**) Ang II concentration. Western blot results (**c**) and corresponding quantification (**d**) of ACE. The WKY group was used as the calibrator with a given value of 100%, and the other groups were compared with this calibrator. *HRS* hydrogen-rich saline; *Ang II* angiotensin II; *ACE* angiotensin converting enzyme; Values are means ± SD. $n = 11$ in each group. *$P < 0.05$ versus WKY; # $P < 0.05$ versus SHR

as in animal models [36, 37]. Moreover, Ang II could induce ROS formation derived from NADPH oxdase, which in turn induced mitochondrial ROS formation and mitochondrial dysfunction [26]. Ang II is also capable of inducing an inflammatory response in the cardiac tissue through the activation of NF-κB [38, 39]. In the current study, HRS treatment suppressed Ang II generation through down-regulating ACE expression in left ventricles of SHR, which interpreted why HRS treatment could suppress NADPH oxidase, preserve mitochondrial function, and inhibit inflammation in left ventricles of SHR, at least in part.

Conclusion

Chronic treatment with HRS attenuates left ventricular hypertrophy in SHR, at least in part, because of its antioxidant, mitochondria-protective, and anti-inflammatory properties.

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