Int J Clin Exp Med 2015;8(2):2506-2515
www.ijcem.com /ISSN:1940-5901/IJCEM0004382

Original Article

The expression and functional evidence for voltage-dependent potassium channel Kv1.3 in lymphocytes during aging in spontaneously hypertensive rats

Ling-Peng Wang[1*], Jian Luo[2*], Hai-Feng Hu[3], Li Zhang[2], Ya-Li Li[2], Li-Man Ai[1], Yu-Ling Wang[2], Yi-Tong Ma[1], Hu-Yati Mu[1], Yue-Mei Hou[1]

[1]Department of Cardiology of The First Affiliated Hospital, Xinjiang Medical University, Urumqi 830000, Xinjiang, China; [2]Department of Internal Medicine (VIP) of The First Affiliated Hospital, Xinjiang Medical University, Urumqi 830000, Xinjiang, China; [3]Department of Heart and Renal of The Sixth People's Hospital in Xinjiang Uygur Autonomous Region, Urumqi 830000, Xinjiang, China. [*]Equal contributors and co-first authors.

Received December 2, 2014; Accepted January 29, 2015; Epub February 15, 2015; Published February 28, 2015

Abstract: Aims: Our previous studies showed that expression and functional profile of voltage-dependent potassium channels Kv1.3 were increased in lymphocytes of spontaneously hypertensive rats (SHR) compared to normotensive rats, suggesting a crucial role for lymphocyte Kv1.3 in the development of hypertension. Here, we further investigated whether the expression and functional profile of Kv1.3 was related to increased blood pressure in SHR with age of 4, 8, 16 and 24 wk. Methods: Systolic blood pressure was measured through pressure device around the tail. mRNA and protein expression were assessed by real-time PCR and western blot in lymphocytes of SHR. Current density of Kv channels in lymphocytes was measured by patch-clamp. Results: Systolic blood pressure was elevated in an age-dependent manner (ANOVA $P < 0.05$). mRNA and protein level of Kv1.3 were significantly increased in an age-dependent manner in lymphocyte of SHR (ANOVA $P < 0.05$). Moreover, the current density of Kv was dramatically enhanced in an age-dependent manner (ANOVA $P < 0.05$). Conclusion: The systolic blood pressure positively correlated with expression as well as current density of potassium channels in lymphocytes of SHR at age of 8, 16 and 24 wk. In conclusion, Kv1.3 channels were upregulated in an age-dependent manner in SHR and correlates with systolic blood pressure during aging. The present study implies that Kv1.3 blockers may be applied as a therapeutic treatment for the development of hypertension during aging.

Keywords: Spontaneously hypertensive rat, lymphocyte, Kv1.3, blood pressure, aging

Introduction

Low-grade inflammation has been recognized as a mechanism contributing to the progression of cardiovascular diseases. More recently, participation of the innate and the adaptive immune response in mechanism that contributes to inflammation has been reported in cardiovascular diseases such as hypertension [1], atherosclerosis [2] and diabetes [3]. Several experimental evidences showed that application of immunosuppressor attenuated development of salt sensitivity [4, 5] and reduced renal pathology in angiotensin II-induced hypertensive mice [6]. However, the detailed insight in this process remains unclear.

The potassium channels expressed by lymphocytes play a critical role in the control of the membrane potential and calcium homeostasis, thereby affecting signal transduction pathways that lead to the activation of these cells following immune response [7]. For instance, activation of voltage dependent potassium channel 1.3 (Kv1.3), which is relatively restricted to the immune system [8], regulates the proliferation, maturation and differentiation of T-cells [9]. On the other hand, Kv1.3 has been observed to be upregulated in pathological conditions such as allergic asthma [10] and type 1 diabetes [11]. Therefore, activation of potassium channel Kv1.3 regulates lymphocyte function and may play a role in pathophysiology of hypertension.

Table 1. The systolic blood pressure in SHR at different ages

Age	SBP (mmHg)	n
4	117 ± 4.0	10
8	154 ± 7.4*	10
16	184 ± 7.6*,†	10
24	215 ± 6.0*,†,‡	10

SHR: Spontaneously hypertensive rats; SBP: Systolic blood pressure. Values are mean ± SEM. *P < 0.05 vs. 4 wk; †P < 0.05 vs. 8 wk; ‡P < 0.05 vs. 16 wk.

In accordance with this concept, previous studies from our laboratory demonstrated that mRNA and protein level as well as the current density of Kv1.3 channel were dramatically elevated in spontaneously hypertensive rats (SHR) as compared to Wistar rats [12]. More recently, similar evidence has been observed in hypertensive patients in Xinjiang Kazakh in which the current density of Kv1.3 was significantly higher [13], suggesting the crucial role of Kv1.3 in development of hypertension found in rodents may also be translated into humans.

Aging is attributed to numerous structural and functional changes in tissues and organs, such as in the vasculature where increased stiffness and vasoconstriction contribute to development of hypertension [14], as well as in the immune system where the alteration in regulation of lymphocyte activity is present [15]. Since the evidence for the changes in Kv channels in lymphocytes which is related to changes in blood pressure during aging progress has not been explored. Consequently, the aim of the present study is to investigate the alteration of Kv1.3 channels at mRNA and protein level, as well as the functional involvement of Kv channels in SHR during aging. Moreover, the association between changes in Kv channel level and blood pressure during aging was assessed.

Methods

Experimental animals

Male SHR used in the present study were purchased from Vital River Beijing (a Charles River company). Rats with age of 4, 8, 16 and 24 weeks entered the study. The animal protocol was approved by The Animal Ethic Committee at the first affiliation of Xinjiang Medical University.

Blood pressure measurement

Conscious rats (n = 40) were placed in a thermo pad with 40°C for 15 min. the blood pressure was measured by BP-6 device (Chengdu Taimeng Co., China) placed around tail artery.

Peripheral lymphocyte isolation

Relatively pure (> 90%) population of lymphocytes from whole blood of mesenteric veins was isolated by differential centrifugation on a Ficoll-Hypaque density gradient, as previously described [12]. Briefly, isolated mononuclear cells were stimulated with phytohaemagglutinin. A monoclonal antibody, which recognizes the CD3 molecule on T-lymphocytes, was used. Cells were then incubated overnight at 37°C in 5% CO_2. Enriched lymphocytes were determined by high-performance liquid chromatography in a balanced salt solution.

Quantitative real-time PCR analysis

Peripheral lymphocytes were used for detection of Kv1.3 mRNA (GenBank: NM-019270). Total RNA was extracted with Trizol Reagent. cDNA was synthesized with iScript Reverse Transcriptase (Bio-Rad). Quantitative real-time PCR (MyIQ, Bio-Rad) was performed with SYBR Green (TaKaRa, Dalian, China). Kv1.3 mRNA levels were expressed relative to the housekeeping gene β-actin (GenBank: NM-031144) as an endogenous control. Primers were designed using Primer 5.0. Kv1.3 (121 bp): sense 5'-GGAAGCTCCGGGAACAAGTG-3'; antisense 5'-TGCCAGCCCATGGATTCTC-3'. β-actin (150 bp): sense 5'-GGAGATTACTGCCCTGGCTCCTA-3'; antisense 5'-GACTCATCGTACTCCTGCTTGCTG-3'.

Western blot analysis

Lymphocytes were isolated. Supernatants were stored at -80°C. Protein was measured using the Bradford dye procedure with bovine serum albumin as a standard (Bio-Rad). Samples (50 μg of total protein) were loaded on slab gels (10% acrylamide; 1 mm thick), separated by sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE), and transferred to nitrocellulose membranes. Protein transfer was confirmed by visualization of pre-stained molecular weight markers (Bio-Rad). Membranes were blocked with 5% nonfat dry milk

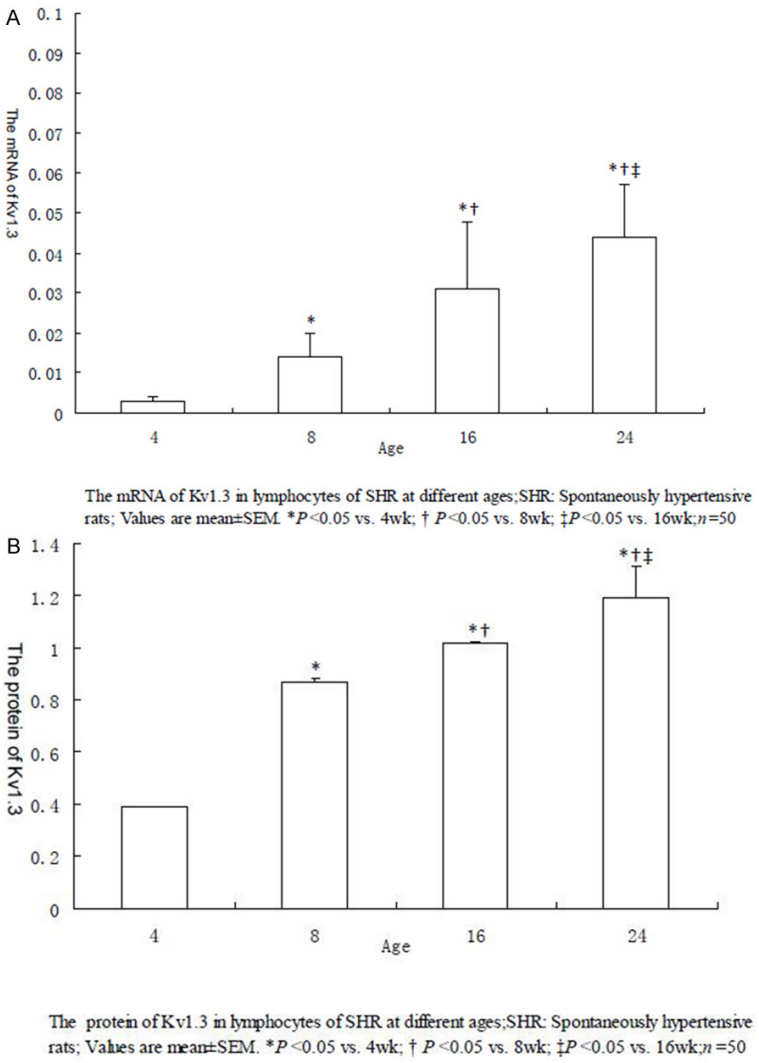


Figure 1. The expression profile of Kv1.3 in lymphocytes of SHR. Shown are mRNA (A) and protein (B) expression of Kv1.3 in lymphocytes in SHR at age of 4, 8, 16 and 24 wk. n = 50 for each age group. Values are mean ± SEM. *P < 0.05 vs. 4 wk, †P < 0.05 vs. 8 wk, ‡P < 0.05 vs. 16 wk.

Table 2. The current density of Kv channels in lymphocytes of SHR at different ages

Age	Density (pA/pF)	n
4	59 ± 7.2	30
8	86 ± 7.9*	30
16	119 ± 10*,†	30
24	126 ± 16*,†,‡	30

SHR: Spontaneously hypertensive rats; SBP: Systolic blood pressure. Values are mean ± SEM. *P < 0.05 vs. 4 wk; †P < 0.05 vs. 8 wk; ‡P < 0.05 vs. 16 wk.

and incubated with primary antibody. The 1:1000 dilutions were used for Kv1.3 (Sigma Aldrich) and β-actin (Santa Cruz). The membranes was stripped and probed for β-actin; this served as an internal control to normalize protein expression. Secondary antibodies (ZSGB-Bio, Beijing) were diluted as 1:1000. Image was analyzed using Quantity-One software (Bio-Rad).

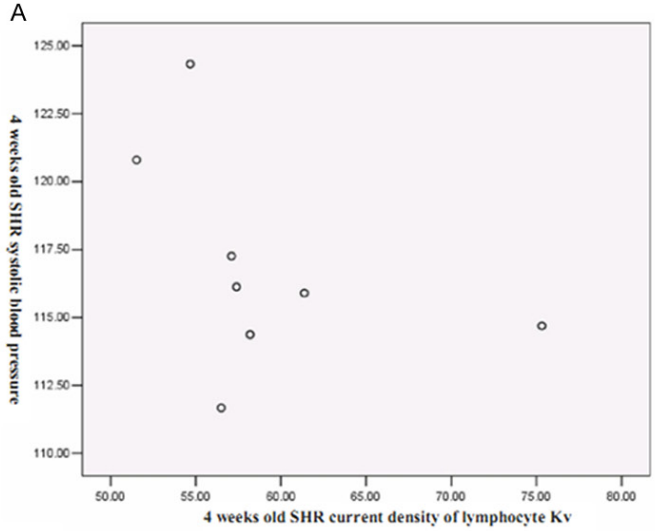
Electrophysiological recording

The whole-cell patch clamp was applied to single lymphocyte using AXON 700B amplifier at room temperature. The external solution was: 150 mM NaCl, 5 mM KCl, 2.5 mM $CaCl_2$, 1.0 mM $MgCl_2$, 10 mM glucose and 10 mM N-(2-hydroxyethyl) piperazine-N'-(2-ethanesulfonic acid) (HEPES); NaOH was added to adjust pH to 7.3. The pipette solution was: 134 mM KCl, 1 mM $CaCl_2$, 10 mM EGTA, 2 mM $MgCl_2$, 10 mM HEPES; KOH was added to adjust pH to 7.3. The concentration free calcium in the internal solution was about 10 nM, assuming the dissociation constant for EGTA. Such a low calcium concentration was applied in order to prevent the activation of calcium-activated potassium channels [12, 16]. The procedure of electrophysiological recordings and data acquisition for whole-cell recording has been reported previously [12, 17]. Currents were measured in voltage-clamp mode and induced by ramp depolarization from -60 mV to +60 mV, 3 s duration, every 15 s with a holding potential of -60 mV. We previously showed the presence of Kv channels in lymphocytes in SHR, and the Kv current can be adequately attenuated by Kv channel blocker 4-aminopyridine (4-AP) (3 mM) [12].

Data analysis

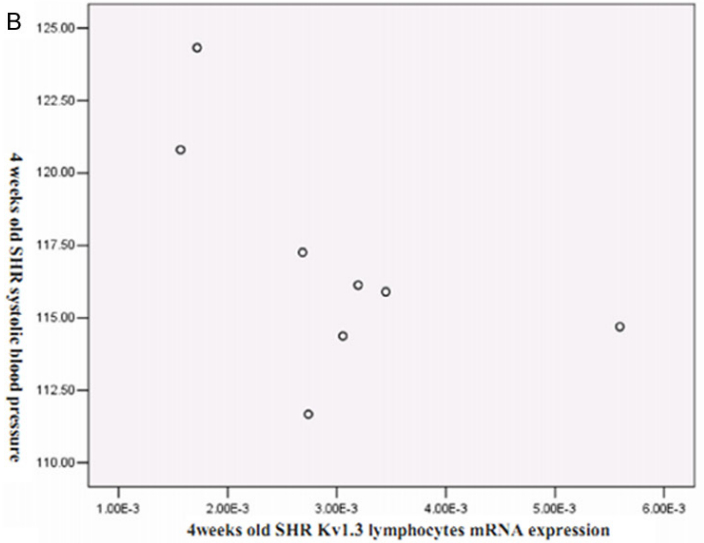
Data are presented as mean ± SEM; statistics were performed using two-way ANOVA, followed by post-hoc using SNK. Statistical significance was accepted when P < 0.05.

A



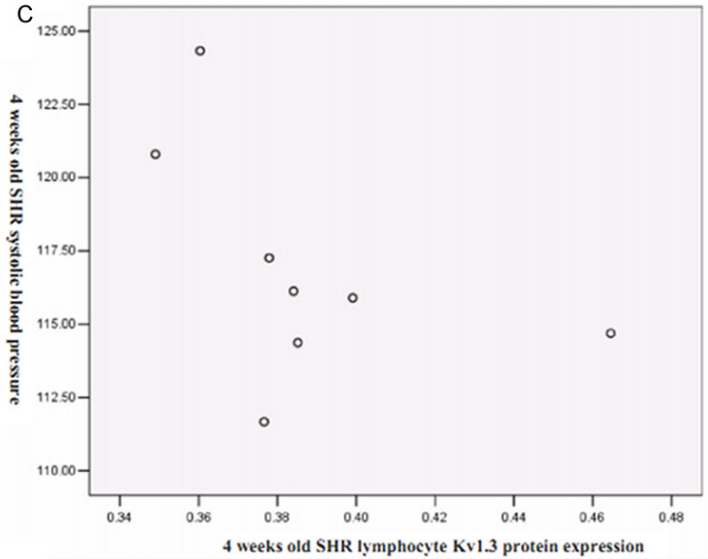
4 weeks old SHR systolic blood pressure and current density of lymphocyte Kv correlation analysis（correlation coefficient $r = 0.421$, $P = 0.299$)

B



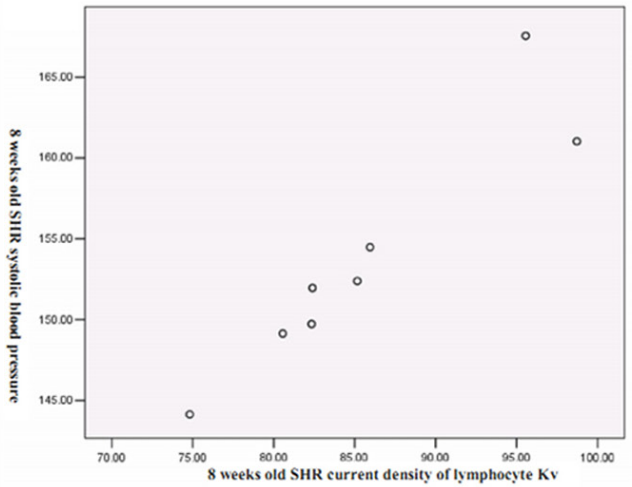
4 weeks old SHR Kv1.3 systolic blood pressure and lymphocytes mRNA expression of correlation analysis（correlation coefficient $r = 0.587$, $P = 0.126$)

C

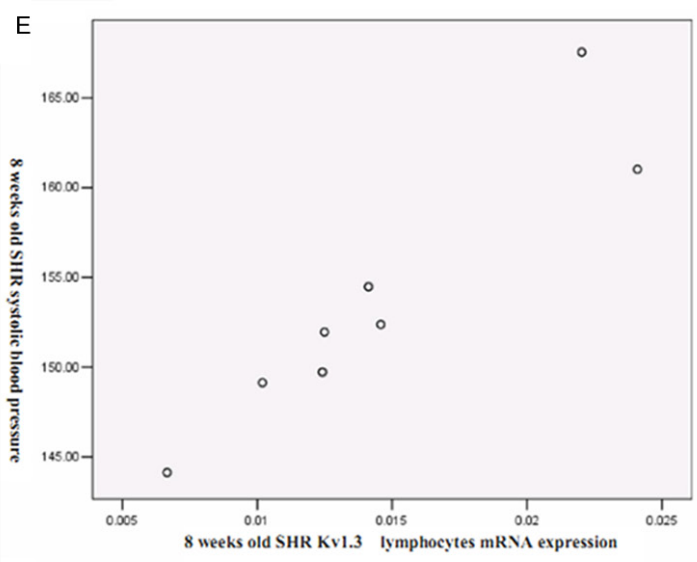


4 weeks old SHR systolic blood pressure and lymphocyte Kv1.3 protein expression of correlation analysis, correlation coefficient r=0.485，P=0.223)

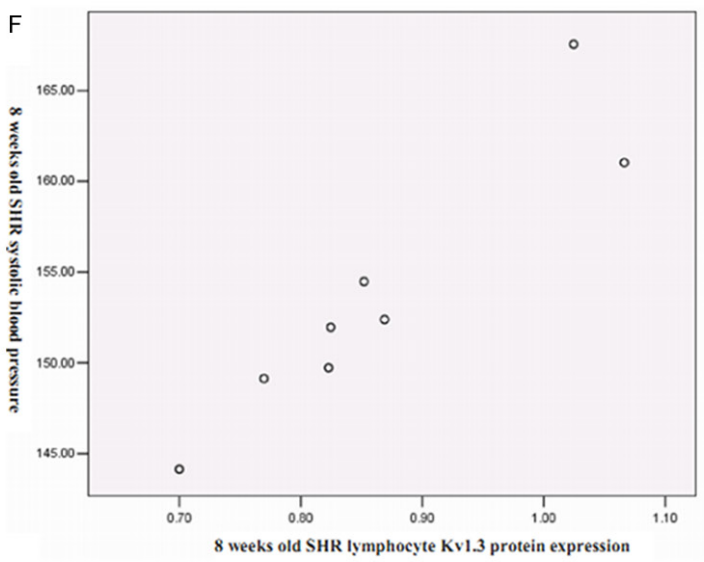
D



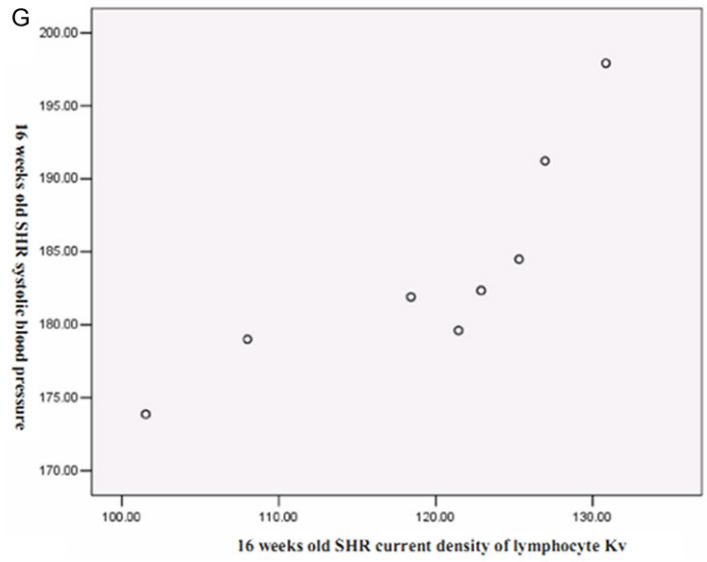
8 weeks old SHR systolic blood pressure and current density of lymphocyte Kv correlation analysis（correlation coefficient r=0.936，P=0.001)



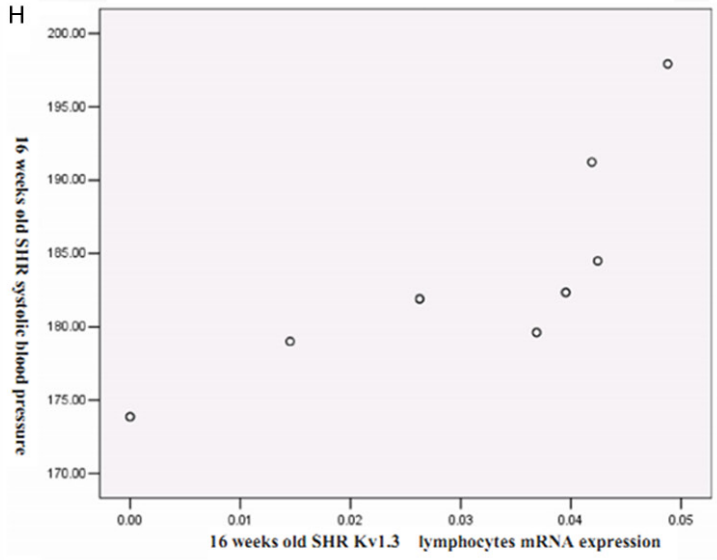
8 weeks old SHR Kv1.3 systolic blood pressure and lymphocytes mRNA expression of correlation analysis（correlation coefficient r=0.936，P=0.01)



8 weeks old SHR systolic blood pressure and lymphocyte Kv1.3 protein expression of correlation analysis, correlation coefficient r=0.935，P=0.001)

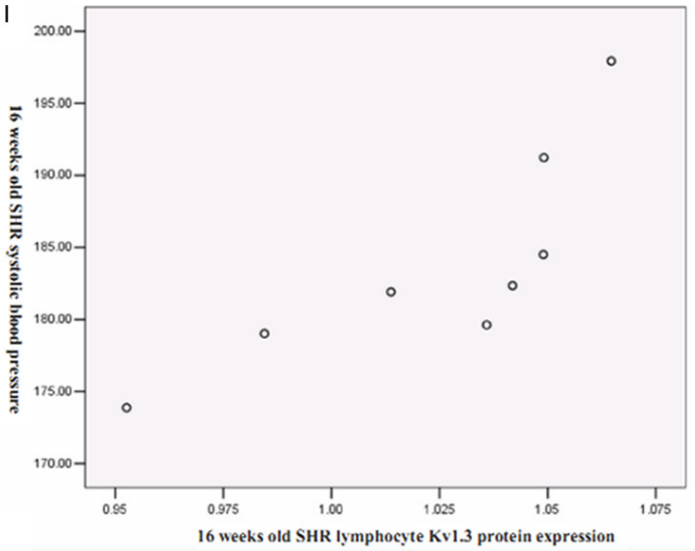


16 weeks old SHR systolic blood pressure and current density of lymphocyte Kv correlation analysis（correlation coefficient r=0.842，P=0.09)



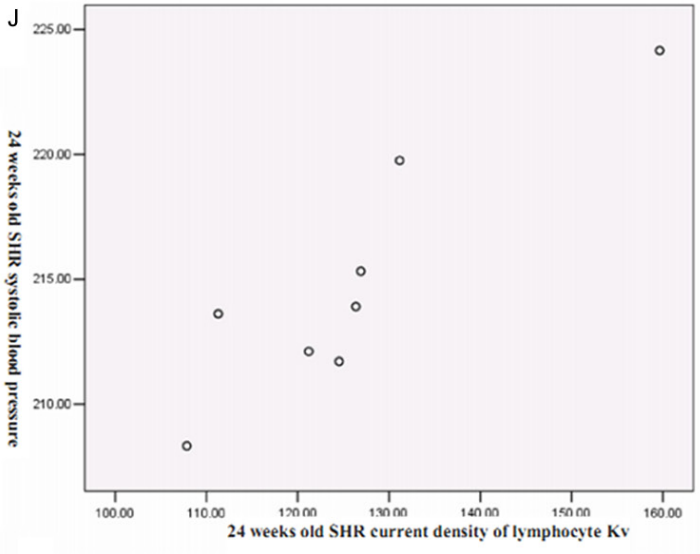
16 weeks old SHR Kv1.3 systolic blood pressure and lymphocytes mRNA expression of correlation analysis（correlation coefficient r=0.796，P=0.018)

I



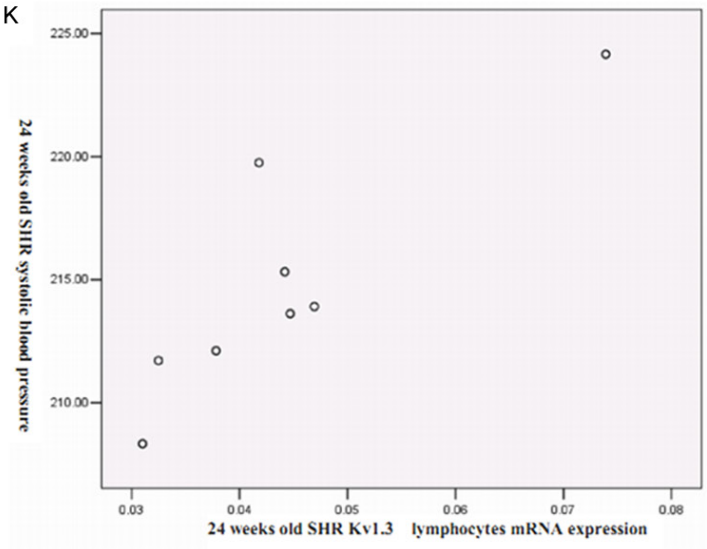
16 weeks old SHR systolic blood pressure and lymphocyte Kv1.3 protein expression of correlation analysis, correlation coefficient r=0.805, P=0.016)

J



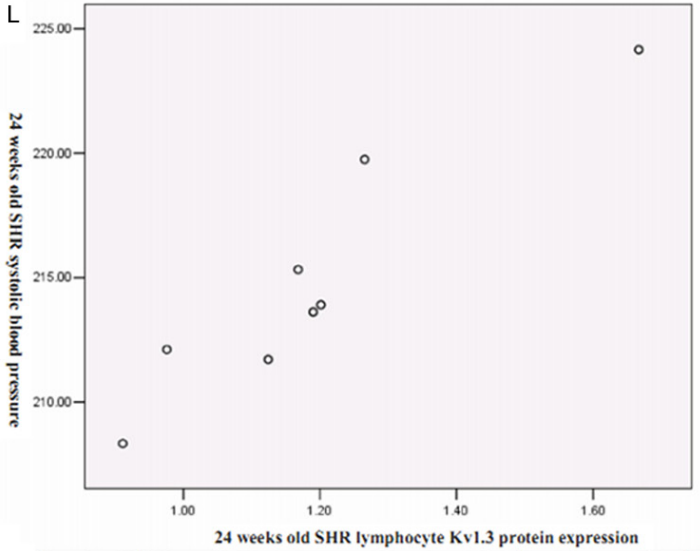
24 weeks old SHR systolic blood pressure and current density of lymphocyte Kv correlation analysis（correlation coefficient r=0.900, P=0.002)

K



24 weeks old SHR Kv1.3 systolic blood pressure and lymphocytes mRNA expression of correlation analysis（correlation coefficient r=0.864, P=0.006)

L



24 weeks old SHR systolic blood pressure and lymphocyte Kv1.3 protein expression of correlation analysis, correlation coefficient r=0.935, P=0.001)

Figure 2. The correlations between systolic blood pressure and expression as well as functional profile of Kv channels in lymphocytes of SHR. Shown are the correlations between systolic blood pressure and current density of Kv at age of 4 wk (A), 8 wk (D), 16 wk (G) and 24 wk (J); between systolic blood pressure and mRNA level of Kv1.3 at age of 4 wk (B), 8 wk (E), 16 wk (H) and 24 wk (K); between systolic blood pressure and protein level of Kv1.3 at age of 4 wk (C), 8 wk (F), 16 wk (I) and 24 wk (L). Values are mean ± SEM.

Results

The alteration of systolic blood pressure in SHR during aging

In accordance with our previous observations [12, 18], the systolic blood pressure of SHR at age of 4 wk was regarded as normal, which was 117 ± 4.0 mmHg in the present study (**Table 1**). The systolic blood pressure of the rest animal groups (with age of 8, 16 and 24 wk) was all elevated as compared to those at age of 4 wk (**Table 1**). Moreover, the systolic blood pressure kept developing during aging, which increased up to 215 ± 5.0 mmHg at age of 24 wk (**Table 1**).

The expression of Kv1.3 in lymphocytes of SHR during aging

The expression of Kv1.3 was minimally present at age of 4 wk, but was significantly increased at all ages in the rest of animals both at mRNA and protein levels (**Figure 1A** and **1B**). Moreover, the mRNA and protein level of Kv1.3 were elevated during aging, which was ~10 fold and ~3 fold higher in animals at age of 24 wk as compared to those at 4 wk, respectively (**Figure 1A** and **1B**).

The alteration of current density of Kv in SHR lymphocytes during aging

In accordance with previous study from our laboratory [18], the Kv current density was 59 ± 7.2 pA/pF in SHR at age of 4 wk (**Table 2**). Notably, the Kv current density was enhanced at all age groups in rest of animals as compared to those at age of 4 wk (**Table 2**). Moreover, the current density kept increasing during aging (at age of 8, 16 and 24 wk), which was more than 2 fold in animals at age of 24 wk as compared to those at age of 4 wk (**Table 2**).

The correlations between systolic blood pressure and Kv current density as well as Kv1.3 expression

There was no significant correlation between systolic blood pressure and Kv current density as well as Kv1.3 expression level in SHR at age of 4 wk (**Figure 2A-C**). However, at all other ages (8, 16 and 24 wk) in SHR, the systolic blood pressure positively correlates with Kv current density (**Figure 2D**, **2G** and **2J**) and Kv1.3 expression (**Figure 2E**, **2H**, **2K**, **2F**, **2I** and **2L**).

Discussion

The main findings of the present study in SHR are 1) the systolic blood pressure was elevated during aging (at age of 4, 8, 16 and 24 wk); 2) the expression of Kv1.3 in lymphocytes was increased at both mRNA and protein level during aging; 3) the current density of Kv was enhanced during aging, and 4) the systolic blood pressure positively correlated with Kv1.3 expression as well as Kv current density at age of 8, 16 and 24 wk, respectively. The implications of these findings are discussed below.

It has been clearly observed and reported in both experimental animals as well as human beings that blood pressure increases with age [19], the latter of which is regarded as one of the risk factors for the development of hypertension [20]. In accordance with one of our previous studies [18] as well as studies from others [21-23], the systolic blood pressure of SHR developed during aging, with almost two-fold increase at age of 24 wk as compared to 4 wk.

Chronic inhibition of the immune system has been shown to attenuate hypertension and renal damage in different animal models of hypertension [24-27] and human patients [28, 29], suggesting activation of immune system, more specifically, immune cells such as lymphocytes contributes to development of hypertension [1]. Since Kv1.3, which is relatively restricted to the immune system [8], plays a critical role in the control of the membrane potential and calcium homeostasis, Kv1.3 contributes to activation of lymphocytes following immune response. Moreover, Kv1.3 has been observed to be upregulated in pathological conditions such as allergic asthma [10] and type 1 diabetes [11]. Therefore, activation of potassium channel Kv1.3 regulates lympho-

cyte activity and may play a role in pathophysiology of hypertension. Indeed, previous studies from our laboratory demonstrated that Kv1.3 mRNA and protein level as well as the current density of Kv channel were dramatically elevated in SHR as compared to Wistar rats [12]. More recently, we have observed that in hypertensive patients in Xinjiang Kazakh in which the current density of Kv channel was significantly higher [13], suggesting the crucial role of Kv1.3 in development of hypertension found in rodents may also be translated into humans. However, little information addresses the changes of Kv1.3 during aging process in lymphocytes, and how this change relates to development of hypertension.

In light of those considerations, we aimed to investigate in the present study the alteration of Kv channels in lymphocytes during aging and assess the association between Kv channels and hypertension. We found that the expression of Kv1.3 at both mRNA and protein levels in SHR was increased during aging. The concept that increased expression of Kv channels during aging is also supported by one of recent studies that Kv4.3 expression was increased in myocytes [30]. Although activation of immune system has been in general regarded to be downregulated during aging in health [15], upregulated Kv1.3 in some diseased states as observed in the present study and studies from others [10, 11] may suggest a role for Kv1.3 in aging process in hypertensive state. In addition to the increased expression of Kv1.3, the current density of Kv channels in lymphocytes of SHR was also enhanced during aging, suggesting a functional evidence for Kv channels in the development of hypertension during aging. More importantly, a positive correlation between systolic blood pressure and Kv1.3 expression as well as Kv current density has been observed, further supporting the idea that increased activity of Kv channels in lymphocytes plays a crucial role in increased systolic blood pressure during aging in SHR. Further studies need to confirm this correlation using specific Kv1.3 blocker or using lymphocyte knockout techniques for Kv1.3 channels.

Study limitations

Our previous studies demonstrated that mRNA and protein level as well as the current density of Kv1.3 channel were dramatically elevated in SHR as compared to Wistar rats [12], suggesting that activation of Kv1.3 channels in lymphocytes plays a crucial role in development of hypertension. In the present study, the expression as well as current density of Kv1.3 was elevated during aging in hypertensive state, suggesting Kv1.3 not only plays a role in hypertension but is also involved in age-associated increased blood pressure. Although the expression and functional profile for Kv1.3 was not assessed in normotensive rats, the immune response in healthy conditions during aging has shown to be dysregulated [15], which may imply that the potassium channel of the immune system is unlikely to be changed or increased.

Conclusions

The present study demonstrates that Kv1.3 channels in lymphocytes are upregulated in SHR during aging, which correlates with increased systolic blood pressure. Together with one of our previous studies that Kv1.3 in lymphocytes is enhanced in SHR as compared to Wistar rats [12], the present study suggests that activation of Kv1.3 channels not only regulates blood pressure, but also plays a role in age-associated hypertension.

Acknowledgements

This study was supported by The Natural Science Fund of Xinjiang Uygur Autonomous Region (2014211C077).

Disclosure of conflict of interest

None.

Address correspondence to: Hu-Yati Mu or Yue-Mei Hou, Department of Cardiology of The First Affiliated Hospital, Xinjiang Medical University, Urumqi 830000, Xinjiang, China. E-mail: muhuyati2014@sina.com

References

[1] Schiffrin EL. Immune mechanisms in hypertension and vascular injury. Clin Sci (Lond) 2014; 126: 267-274.

[2] Ait-Oufella H, Sage AP, Mallat Z and Tedgui A. Adaptive (T and B cells) immunity and control by dendritic cells in atherosclerosis. Circulation Res 2014; 114: 1640-1660.

[3] Li M, Song LJ and Qin XY. Advances in the cellular immunological pathogenesis of type 1 diabetes. J Cell Mol Med 2014; 18: 749-758.

[4] Quiroz Y, Pons H, Gordon KL, Rincon J, Chavez M, Parra G, Herrera-Acosta J, Gomez-Garre D, Largo R, Egido J, Johnson RJ and Rodriguez-Iturbe B. Mycophenolate mofetil prevents salt-sensitive hypertension resulting from nitric oxide synthesis inhibition. Am J Physiol Renal Physiol 2001; 281: F38-47.

[5] Rodriguez-Iturbe B, Pons H, Quiroz Y, Gordon K, Rincon J, Chavez M, Parra G, Herrera-Acosta J, Gomez-Garre D, Largo R, Egido J and Johnson RJ. Mycophenolate mofetil prevents salt-sensitive hypertension resulting from angiotensin II exposure. Kidney Int 2001; 59: 2222-2232.

[6] Crowley SD, Frey CW, Gould SK, Griffiths R, Ruiz P, Burchette JL, Howell DN, Makhanova N, Yan M, Kim HS, Tharaux PL and Coffman TM. Stimulation of lymphocyte responses by angiotensin II promotes kidney injury in hypertension. Am J Physiol Renal Physiol 2008; 295: F515-524.

[7] Feske S, Skolnik EY and Prakriya M. Ion channels and transporters in lymphocyte function and immunity. Nat Rev Immunol 2012; 12: 532-547.

[8] Lewis RS and Cahalan MD. Subset-specific expression of potassium channels in developing murine T lymphocytes. Science 1988; 239: 771-775.

[9] Panyi G, Varga Z and Gaspar R. Ion channels and lymphocyte activation. Immunol Lett 2004; 92: 55-66.

[10] Koshy S, Huq R, Tanner MR, Atik MA, Porter PC, Khan FS, Pennington MW, Hanania NA, Corry DB and Beeton C. Blocking KV1.3 channels inhibits Th2 lymphocyte function and treats a rat model of asthma. J Biol Chem 2014; 289: 12623-12632.

[11] Toldi G, Vasarhelyi B, Kaposi A, Meszaros G, Panczel P, Hosszufalusi N, Tulassay T and Treszl A. Lymphocyte activation in type 1 diabetes mellitus: the increased significance of Kv1.3 potassium channels. Immunol Lett 2010; 133: 35-41.

[12] Luo J, Zhang YM, Ma KT, Si JQ and Liang P. [Difference in the expression of Kv channel in lymphocytes between spontaneously hypertensive rats and Wistar rats]. Sheng Li Xue Bao 2010; 62: 382-386.

[13] Zhang QB, Zhang YM, Cheng LF, Yuan QY, Zhang GM, Liang P and Gou F. [Voltage-dependent potassium channel and calcium-activated potassium channel current changes of peripheral blood T-lymphocytes from hypertensive patients in Xinjiang Kazakh]. Zhonghua Xin Xue Guan Bing Za Zhi 2013; 41: 1020-1024.

[14] Denker MG and Cohen DL. What is an appropriate blood pressure goal for the elderly: review of recent studies and practical recommendations. Clin Interv Aging 2013; 8: 1505-1517.

[15] Fulop T, Le Page A, Fortin C, Witkowski JM, Dupuis G and Larbi A. Cellular signaling in the aging immune system. Curr Opin Immunol 2014; 29: 105-111.

[16] Grissmer S, Nguyen AN and Cahalan MD. Calcium-activated potassium channels in resting and activated human T lymphocytes. Expression levels, calcium dependence, ion selectivity, and pharmacology. J Gen Physiol 1993; 102: 601-630.

[17] Ohya S, Nakamura E, Horiba S, Kito H, Matsui M, Yamamura H and Imaizumi Y. Role of the K(Ca)3.1 K+ channel in auricular lymph node CD4+ T-lymphocyte function of the delayed-type hypersensitivity model. Br J Pharmacol 2013; 169: 1011-1023.

[18] Luo J, Ma KT, Zhang YM, Si JQ, Liang P and Li J. [Effects of telmisartan on 4-Aminopyridine-sensitive voltage dependant potassium channel of lymphocyte derived from spontaneously hypertensive rat]. Zhonghua Xin Xue Guan Bing Za Zhi 2010; 38: 751-754.

[19] Edwards EW, DiPette DJ, Townsend RR and Cohen DL. Top 10 landmark studies in hypertension. J Am Soc Hypertens 2014; 8: 437-447.

[20] Turgut F, Yesil Y, Balogun RA and Abdel-Rahman EM. Hypertension in the elderly: unique challenges and management. Clin Geriatr Med 2013; 29: 593-609.

[21] Rossoni LV, Oliveira RA, Caffaro RR, Miana M, Sanz-Rosa D, Koike MK, Do Amaral SL, Michelini LC, Lahera V and Cachofeiro V. Cardiac benefits of exercise training in aging spontaneously hypertensive rats. J Hypertens 2011; 29: 2349-2358.

[22] Scridon A, Gallet C, Arisha MM, Orea V, Chapuis B, Li N, Tabib A, Christe G, Barres C, Julien C and Chevalier P. Unprovoked atrial tachyarrhythmias in aging spontaneously hypertensive rats: the role of the autonomic nervous system. Am J Physiol Heart Circ Physiol 2012; 303: H386-392.

[23] Sun MW, Qian FL, Wang J, Tao T, Guo J, Wang L, Lu AY and Chen H. Low-intensity voluntary running lowers blood pressure with simultaneous improvement in endothelium-dependent vasodilatation and insulin sensitivity in aged spontaneously hypertensive rats. Hypertens Res 2008; 31: 543-552.

[24] De Miguel C, Guo C, Lund H, Feng D and Mattson DL. Infiltrating T lymphocytes in the kidney increase oxidative stress and participate in the development of hypertension and renal disease. Am J Physiol Renal Physiol 2011; 300: F734-742.

[25] Mattson DL, James L, Berdan EA and Meister CJ. Immune suppression attenuates hyperten-

sion and renal disease in the Dahl salt-sensitive rat. Hypertens 2006; 48: 149-156.

[26] Muller DN, Shagdarsuren E, Park JK, Dechend R, Mervaala E, Hampich F, Fiebeler A, Ju X, Finckenberg P, Theuer J, Viedt C, Kreuzer J, Heidecke H, Haller H, Zenke M and Luft FC. Immunosuppressive treatment protects against angiotensin II-induced renal damage. Am J Pathol 2002; 161: 1679-1693.

[27] Pechman KR, Basile DP, Lund H and Mattson DL. Immune suppression blocks sodium-sensitive hypertension following recovery from ischemic acute renal failure. Am J Physiol Regul Integr Comp Physiol 2008; 294: R1234-1239.

[28] Herrera J, Ferrebuz A, MacGregor EG and Rodriguez-Iturbe B. Mycophenolate mofetil treatment improves hypertension in patients with psoriasis and rheumatoid arthritis. J Am Soc Nephrol 2006; 17: S218-225.

[29] Seaberg EC, Munoz A, Lu M, Detels R, Margolick JB, Riddler SA, Williams CM and Phair JP. Association between highly active antiretroviral therapy and hypertension in a large cohort of men followed from 1984 to 2003. AIDS 2005; 19: 953-960.

[30] Plotnikov AN, Sosunov EA, Patberg KW, Anyukhovsky EP, Gainullin RZ, Shlapakova IN, Krishnamurthy G, Danilo P Jr. and Rosen MR. Cardiac memory evolves with age in association with development of the transient outward current. Circulation 2004; 110: 489-495.