Bradykinin β_2 Receptor –58T/C Gene Polymorphism and Essential Hypertension: A Meta-Analysis

Yan-yan Li^{1*}, Hui Zhang², Jian Xu¹, Yun Qian¹, Xin-zheng Lu², Bing Yang¹, Minglong Chen¹, Zhi-jian Yang^{1,2}, Ke-jiang Cao^{2*}

1 Department of Geriatrics, First Affiliated Hospital of Nanjing Medical University, Nanjing, China, **2** Department of Cardiology, First Affiliated Hospital of Nanjing Medical University, Nanjing, China

Abstract

Background: Research has shown that bradykinin β_2 receptor (*BDKRB2*) –58T/C gene polymorphism is correlated with the risk of essential hypertension (EH), but the results remain inconclusive.

Objective and Methods: The objective of this study was to explore the association between *BDKRB2*–58T/C gene polymorphism and EH. A meta-analysis of 11 studies with 3882 subjects was conducted. Pooled odds ratios (ORs) for the association between *BDKRB2*–58T/C gene polymorphism and EH and their corresponding 95% confidence intervals (CIs) were estimated using the random effects model.

Results: The *BDKRB2*–58T/C gene polymorphism was significantly correlated with EH under an allelic genetic model (OR = 1.24, 95% CI = 1.05–1.46; $P = 0.01$), a dominant genetic model (OR = 0.65, 95% CI = 0.47–0.90; $P = 0.01$), a recessive genetic model (OR = 1.146, 95% CI = 1.035–1.269; $P = 0.009$), a homozygote genetic model (OR = 1.134, 95% CI = 1.048–1.228; $P = 0.002$), and a heterozygote genetic model (OR = 1.060, 95% CI = 1.009–1.112; $P = 0.019$).

Conclusions: The *BDKRB2*–58T/C gene polymorphism is associated with increased EH risk. The results of this study suggest that carriers of the –58C allele are susceptible to EH.

Citation: Li Y-y, Zhang H, Xu J, Qian Y, Lu X-z, et al. (2012) Bradykinin β_2 Receptor –58T/C Gene Polymorphism and Essential Hypertension: A Meta-Analysis. PLoS ONE 7(8): e43068. doi:10.1371/journal.pone.0043068

Editor: Yuri E. Khudyakov, Centers for Disease Control and Prevention, United States of America

Received: May 1, 2012; **Accepted:** July 16, 2012; **Published:** August 10, 2012

Copyright: © 2012 Li et al. This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

Funding: This work was funded by the National Natural Science Foundation of China (NSFC 81100073 to Dr Yan-yan Li) and the Priority Academic Program Development of Jiangsu Higher Education Institutions (PAPD). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Competing Interests: The authors have declared that no competing interests exist.

* E-mail: liynjmu123@126.com (YL); kjcao@njmu.edu.cn (KC)

Introduction

The kallikrein–kinin system (KKS) is an important hormonal system that takes part in blood pressure (BP) and renal sodium regulation [1]. Bradykinin is one of the strongest vasodilator substances, and it has powerful diuretic effects. It exerts its vasodilatory effects mainly by expanding the vessels directly, confronting the vasoconstrictive effects of angiotensin II and noradrenaline, and promoting the synthesis of endogenous vasodilator substances, such as nitric oxide.

The function of bradykinin is mediated by two receptor subtypes, namely, β_1 (*BDKRB1*) and β_2 (*BDKRB2*) [2]. *BDKRB1* expression is low in healthy individuals; however, under such pathological states as inflammation and tissue injury, it could be upregulated. The physical effects of bradykinin are mostly mediated by *BDKRB2*. The *BDKRB2* gene, located in 14q32.1–32.2, spans approximately 4 kb and consists of three exons. In 1996, Braun et al. detected –58T/C variations in the *BDKRB2* promoter region and found that these could lead to a reduction in *BDKRB2* transcription, which might be associated with the pathogenesis of essential hypertension (EH) [3].

The relationship between *BDKRB2*–58T/C gene polymorphism and EH has been widely studied, but the results remain

inconclusive. In 2000, Gainer et al. found that *BDKRB2*–58C might represent a susceptibility marker for EH in African Americans [4]. In 2006, Dong et al. found an association between *BDKRB2*–58T/C gene polymorphism and EH and concluded that the –58CC genotype is associated with increased EH risk in a Chinese population [5]. However, in 2012, Bhupatiraju et al. did not identify any such association in an Indian population [6]. We thus performed a meta-analysis of 11 studies, including 1947 patients with EH and 1935 control subjects, to deduce a reasonable conclusion on the relationship between *BDKRB2*–58T/C gene polymorphism and EH (Supplement S5).

Materials and Methods

Publication Search and Inclusion Criteria

PubMed, EMBASE, Web of Science, China Biological Medicine Database, and China National Knowledge Infrastructure were searched for relevant articles with the terms “essential hypertension”, “bradykinin β_2 receptor”, and “polymorphism”. Studies published before 2012 were obtained (last research updated on July 7, 2012).

Review: Bradykinin receptor B2 -58T/C gene polymorphism and essential hypertension (Version 01)
 Comparison: 01 EH group versus control group
 Outcome: 02 Distribution of C allelic frequency of Bradykinin receptor B2 -58T/C gene polymorphism

Figure 1. Forest plot of essential hypertension associated with *BDKRB2* -58T/C gene polymorphism under an allelic genetic model (distribution of C allelic frequency of *BDKRB2* gene).

doi:10.1371/journal.pone.0043068.g001

Studies that met the following major criteria were included: (a) the association between *BDKRB2*-58T/C gene polymorphism and EH was assessed; (b) the diagnosis of EH was in line with the 1999 diagnostic criteria of the World Health Organization in which systolic BP ≥ 140 mmHg, diastolic BP ≥ 90 mmHg, and treatment with antihypertensive medication defined EH, excluding patients with secondary hypertension, cardiomyopathy, valvular heart disease, congenital heart diseases, and renal failure; (c) the Hardy-Weinberg equilibrium (HWE) was followed; and (d) the results of the same data used in different studies were adopted only once. If articles with similar data were published by the same work group, the study with the larger sample size was selected.

Data Extraction

The data were collected according to a standard protocol. Repeated publications, studies against the selection criteria, and work providing insufficient data were excluded from the meta-analysis. Data were recorded as follows: first author's name, publication year, study region, number of genotypes, genotyping, study design, matching criteria, total number of case patients, and total number of control subjects.

Statistical Analysis

Five genetic models (allelic, dominant, recessive, homozygote, and heterozygote) were used. The association between *BDKRB2*-58T/C gene polymorphism and EH reported under these models was analyzed using odds ratios (ORs) with 95% confidence intervals (CIs). Between-study heterogeneity was calculated by χ^2 -based Q analysis, and significance was set at $P < 0.05$ [7]. The variation caused by heterogeneity was estimated

by calculating the inconsistency index I^2 . If heterogeneity among studies was detected, the DerSimonian-Laird random effects pooling method was used [8]; otherwise, the Mantel-Haenszel method fixed effects model was applied [9]. The Z test was used to determine the pooled OR, and significance was set at $P < 0.05$.

Fisher's exact test was used to assess the HWE, and $P < 0.05$ was considered statistically significant. Potential publication bias was estimated by funnel plot analysis. The funnel plot asymmetry was assessed by Egger's linear regression test on the natural logarithm scale of the OR ($P < 0.05$) [10]. Statistical analysis was performed using STATA 11.0 (StataCorp, College Station, TX).

Results

Studies and Populations

Our literature search yielded 20 relevant articles, 11 of which met the inclusion criteria. Of the 9 excluded studies, 2 were double publications, 2 were reviews, and 5 were not associated with the *BDKRB2*-58T/C gene polymorphism. No study was excluded for deviating from the HWE. Data were collected from 1947 patients with EH and 1935 control subjects (Supplement S1 and S6) [4–6,11–18]. The six surveyed regions representing Asia, North America, and Europe included China, Japan, India, the United States, Canada, and Italy.

Pooled Analysis

A significant association between *BDKRB2*-58T/C gene polymorphism and EH was found under the allelic (OR = 1.24, 95% CI = 1.05–1.46; $P = 0.01$), dominant (OR = 0.65, 95% CI = 0.47–0.90; $P = 0.01$), recessive (OR = 1.146, 95% CI = 1.035–1.269;

Review: Bradykinin receptor B2 -58T/C gene polymorphism and essential hypertension (Version 01)
 Comparison: 01 EH group versus control group
 Outcome: 01 TT vs. TC+CC

Figure 2. Forest plot of essential hypertension associated with BDKRB2 -58T/C gene polymorphism under a dominant genetic model (TT/TC+CC).

doi:10.1371/journal.pone.0043068.g002

Review: Bradykinin receptor B2 -58T/C gene polymorphism and essential hypertension
 Comparison: 01 EH group versus control group
 Outcome: 02 Distribution of C allelic frequency of Bradykinin receptor B2 -58T/C gene polymorphism

Figure 3. Forest plot of essential hypertension associated with BDKRB2 -58T/C gene polymorphism under an allelic genetic model stratified by TT0 TT0: TT genotype sample size of control group.

doi:10.1371/journal.pone.0043068.g003

Review: Bradykinin receptor B2 -58T/C gene polymorphism and essential hypertension
 Comparison: 01 EH group versus control group
 Outcome: 01 TT vs. TC+CC

Figure 4. Forest plot of essential hypertension associated with *BDKRB2*-58T/C gene polymorphism under a dominant genetic model stratified by region.

doi:10.1371/journal.pone.0043068.g004

$P = 0.009$), homozygote (OR = 1.134, 95% CI = 1.048–1.228; $P = 0.002$), and heterozygote (OR = 1.060, 95% CI = 1.009–1.112; $P = 0.019$) genetic models. Subgroup analysis stratified by continent also revealed a significant association between *BDKRB2*-58T/C gene polymorphism and EH in the Asian subgroup under the allelic (OR = 1.24, 95% CI = 1.01–1.52; $P = 0.04$), dominant (OR = 0.63, 95% CI = 0.43–0.92; $P = 0.02$), recessive (OR = 1.152, 95% CI = 1.010–1.315; $P = 0.036$), homozygote (OR = 1.166, 95% CI = 1.047–1.298; $P = 0.005$), and heterozygote (OR = 1.070, 95% CI = 1.011–1.132; $P = 0.019$) genetic models. No significant association was found in the American and European subgroups under any of the genetic models ($P > 0.05$) (Supplement S2; Figures 1 and 2).

In consideration of the significant heterogeneity we observed, a meta-regression was performed to explore the source of heterogeneity. Under the allelic genetic model, the heterogeneity could be explained by the number of control subjects with the TT genotype (TT0) ($P = 0.036$), total sample size of the control group ($P = 0.037$), and study region ($P = 0.045$). Based on TT0, the whole population was divided into two subgroups: Subgroup 1 was

defined as TT0 < 30, whereas subgroup 2 was classified as TT0 > 30. In the subgroup analysis by TT0, under the allelic genetic model, significantly increased EH risk was detected in subgroup 1 (OR = 1.42, 95% CI = 1.08–1.87; $P = 0.01$, $P_{\text{heterogeneity}} = 0.02$). In subgroup 2, no significant increase in EH risk was found (OR = 1.09, 95% CI = 1.05–1.46; $P = 0.33$, $P_{\text{heterogeneity}} = 0.05$) (Supplement S2 and S3; Figure 3).

Under the dominant genetic model, the heterogeneity could be explained by the study region ($P = 0.011$), total sample size of the control group ($P = 0.033$), and TT0 ($P = 0.041$). The entire population was separated into six subgroups by study region (China, Japan, the United States, Italy, India, and Canada). A significant association was found in the Canadian subgroup (OR = 0.14, 95% CI = 0.03–0.62; $P = 0.01$), but not in the other five subgroups ($P > 0.05$) (Supplement S2 and S4; Figure 4).

Bias Diagnostics

Publication bias was assessed using funnel plot analysis and Egger's test. The funnel plot did not show evidence of publication bias (Figure 5). Similarly, the absence of statistically significant

Figure 5. Funnel plot for studies of the association of essential hypertension associated with *BDKRB2* -58T/C gene polymorphism under an allelic genetic model (distribution of C allelic frequency of *BDKRB2* gene). The horizontal and vertical axis correspond to the OR and confidence limits. OR: odds ratio; SE: standard error.

doi:10.1371/journal.pone.0043068.g005

differences in Egger's test indicated that there was no publication bias in the current meta-analysis under the allelic genetic model ($T = -0.78$; $P = 0.455$).

Discussion

This meta-analysis found a significant association between *BDKRB2*-58T/C gene polymorphism and EH: OR = 1.24 for the allelic genetic model, OR = 0.65 for the dominant genetic model, OR = 1.146 for the recessive genetic model, OR = 1.134 for the homozygote genetic model, and OR = 1.060 for the heterozygote genetic model. The subgroup analysis stratified by continent also revealed that the *BDKRB2*-58T/C gene polymorphism was significantly correlated with EH in the Asian subgroup ($P < 0.05$), but not in the American and European subgroups ($P > 0.05$). Such variation in the results can be attributed to the ethnic differences between groups.

In the following meta-regression to explore the heterogeneity source under an allelic genetic model, the confounding factors TT0, study region, and total sample size of the control group could partly explain the heterogeneity source, with TT0 being the most important factor. This finding suggested that the heterogeneity among the different studies can be ascribed to the nonuniformity in the sample size of control subjects with the TT genotype. Subgroup analysis according to the number of control subjects with the TT genotype revealed that EH risk was significantly increased only in the TT0<30 subgroup.

Research has shown that the *BDKRB2*-58T/C gene polymorphism is associated with increased EH risk, especially in Asians. Carriers of the -58C allele of the *BDKRB2* gene might be predisposed to developing EH. In 2010, Niu et al. performed a meta-analysis aiming to provide a comprehensive evaluation of the correlation between *BDKRB2*-58T/C gene polymorphism and EH. They found that the -58T allele exhibited a protective effect on hypertension only in African Americans ($P = 0.04$) and had no effect on hypertension in Asians and Caucasians ($P > 0.05$) [19]. Their results differ from those we obtained in the present meta-analysis. Niu et al. included only 4 studies with 823 case patients and 916 control subjects, whereas we analyzed 11 studies with 1945 case patients and 1937 control subjects. Therefore, our results should be more reasonable than theirs.

EH is a polygenic disease caused by both environmental and hereditary factors [20]. Research has shown that the KKS plays an important role in regulating BP. As a vasoactive substance, bradykinin could adjust the release of a series of biological active media, such as prostaglandin, nitric oxide, and platelet-activating factor, thus causing vessels to dilate and the BP to drop and suppress the proliferation of smooth muscle cells, which exert a protective effect on the cardiovascular system [14]. *BDKRB2* is a transmembrane G protein-coupled protein. It generally mediates most functions of the KKS. An abnormal gene structure of *BDKRB2* would contribute to EH and other cardiovascular diseases, suggesting that it is a candidate gene for EH. Bradykinin could lower renal vascular resistance and increase the renal blood flow volume, which in turn could increase urine sodium excretion [21–22]. *BDKRB2*-58T/C mutation could lower the transcription rate of the *BDKRB2* gene, constrict vessels, and create an imbalance between water and salt, which all contribute to EH. The results of the current meta-analysis have confirmed these findings.

This work was not without limitations. The large-scale studies on the relationship between EH and *BDKRB2*-58T/C gene polymorphism included in the meta-analysis still proved inadequate. *BDKRB2* was affected not only by the *BDKRB2*-58T/C gene polymorphism but also by such environmental factors as tissue injury and inflammation. Lastly, the type of association between *BDKRB2*-58T/C gene polymorphism and EH could not be easily adjusted. All these represent a weakness of the study.

The results of this meta-analysis suggest that the *BDKRB2*-58C allele might increase EH risk. This conclusion provides a strong foundation for formulating a strategy for individual therapy in patients with EH. Given the abovementioned limitations, however, this finding warrants further investigation.

Supporting Information

Supplement S1 Characteristics of the investigated studies of the association between bradykinin β 2 receptor -58T/C gene polymorphism and essential hypertension. (DOC)

Supplement S2 Summary of meta-analysis of association of bradykinin β 2 receptor -58T/C gene polymorphism and essential hypertension. (DOC)

Supplement S3 The meta-regression results among 11 studies under an allelic genetic model for -58T/C gene polymorphism of bradykinin β 2 receptor. (DOC)

Supplement S4 The meta-regression results among 11 studies under a dominant genetic model for -58T/C gene polymorphism of bradykinin β 2 receptor. (DOC)

Supplement S5 PRISMA 2009 Checklist. (DOC)

Supplement S6 PRISMA 2009 Flow Diagram. (DOC)

Acknowledgments

Thank all our colleagues working in the Department of geriatrics, the First Affiliated Hospital of Nanjing Medical University.

Author Contributions

Conceived and designed the experiments: YL KC. Performed the experiments: YL HZ. Analyzed the data: YL JX ZY. Contributed

reagents/materials/analysis tools: YL YQ MC. Wrote the paper: YL XL BY.

References

1. Katori M, Majima M (1996) Pivotal role of renal kallikrein-kinin system in the development of hypertension and approaches to new drugs based on this relationship. *Jpn J Pharmacol* 70: 95–128.
2. Braun A, Maier E, Kammerer S, Müller B, Roscher AA (1996) A novel sequence polymorphism in the promoter region of the human B2-bradykinin receptor gene. *Hum Genet* 97: 688–689.
3. Braun A, Kammerer S, Maier E, Böhme E, Roscher AA (1996) Polymorphisms in the gene for the human B2-bradykinin receptor. New tools in assessing a genetic risk for bradykinin-associated diseases. *Immunopharmacology* 33: 32–35.
4. Gainer JV, Brown NJ, Bachvarova M, Bastien L, Maltais I, et al. (2000) Altered frequency of a promoter polymorphism of the kinin B2 receptor gene in hypertensive African-Americans. *Am J Hypertens* 13: 1268–1273.
5. Dong HY, Li QR, Wang Q, Luo ZG (2006) Association of genetic polymorphism of β 2-bradykinin receptor gene and AGT gene with essential hypertension in Shenzhen population. *Acta Academiae Medicinae Militaris Tertiae* 28: 1252–1254.
6. Bhupatiraju C, Patkar S, Pandharpurkar D, Joshi S, Tirunilai P (2012) Association and Interaction of -58C>T and $\pm$ 9 bp Polymorphisms of BDKRB2 Gene Causing Susceptibility to Essential Hypertension. *Clin Exp Hypertens* 34: 230–235.
7. Cochran WG (1968) The effectiveness of adjustment by subclassification in removing bias in observational studies. *Biometrics* 24: 295–313.
8. Mantel N, Haenszel W (1959) Statistical aspects of the analysis of data from retrospective studies of disease. *J Natl Cancer Inst* 22: 719–748.
9. DerSimonian R, Laird N (1986) Meta-analysis in clinical trials. *Control Clin Trials* 7: 177–188.
10. Egger M, Davey Smith G, Schneider M, Minder C (1997) Bias in meta-analysis detected by a simple, graphical test. *British Medical Journal* 315: 629–634.
11. Mukae S, Aoki S, Itoh S, Nishio K, Iwata T, et al. (1999) Promoter polymorphism of the beta2 bradykinin receptor gene is associated with essential hypertension. *Jpn Circ J* 63: 759–762.
12. Aoki S, Mukae S, Itoh S, Sato R, Nishio K, et al. (2001) The genetic factor in acute myocardial infarction with hypertension. *Jpn Circ J* 65: 621–626.
13. Wang B, Dang A, Liu G (2001) Genetic variation in the promoter region of the beta2 bradykinin receptor gene is associated with essential hypertension in a Chinese Han population. *Hypertens Res* 24: 299–302.
14. Fu Y, Katsuya T, Matsuo A, Yamamoto K, Akasaka H, et al. (2004) Relationship of bradykinin B2 receptor gene polymorphism with essential hypertension and left ventricular hypertrophy. *Hypertens Res* 27: 933–938.
15. Li NF, Wang YC, Zhou L, Li T, Wang XL, et al. (2008) Association of genetic variants in the promoter region of the β 2-bradykinin receptor gene and essential hypertension in Kazakans of Xinjiang. *Journal of Medical Molecular Biology* 5: 110–113.
16. Zou L, Li FQ, Zhou P, Chen DL (2011) Relationship between plasma kallikrein B1 and bradykinin receptor B2 gene polymorphism, body mass index and geriatric essential hypertension. *Chinese Journal of Gerontology* 31: 2609–2612.
17. Cui J, Melista E, Chazaro I, Zhang Y, Zhou X, et al. (2005) Sequence variation of bradykinin receptors B1 and B2 and association with hypertension. *J Hypertens* 23: 55–62.
18. Milan A, Mulatero P, Williams TA, Carra R, Schiavone D, et al. (2005) Bradykinin B2 receptor gene (-58T/C) polymorphism influences baroreflex sensitivity in never-treated hypertensive patients. *J Hypertens* 23: 63–69.
19. Niu W, Qi Y, Gao P, Zhu D (2010) A meta-analysis of the bradykinin B2 receptor gene -58C/T polymorphism with hypertension. *Clin Chim Acta* 411: 324–328.
20. Li YY (2012) α -Adducin Gly460Trp gene mutation and essential hypertension in a Chinese population: a meta-analysis including 10,960 subjects. *PLoS One* 7: e30214.
21. Siragy HM (1993) Evidence that intrarenal bradykinin plays a role in regulation of renal function. *Am J Physiol* 265: E648–E654.
22. Alfie ME, Sigmon DH, Pomposiello SI, Carretero OA (1997) Effect of high salt intake in mutant mice lacking bradykinin-B2 receptors. *Hypertension* 29: 483–487.