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Original Article

Association between serum 25-hydroxyvitamin d and early recurrent ischemic stroke

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Abstract

Aim: Vitamin D deficiency has been reported as a new risk factor for ischemic stroke in recent years. However, the pathophysiological mechanisms between recurrent ischemic stroke and decreased 25-hydroxyvitamin D [25(OH) D] are not clear. The aim of this study is to examine the relationship between initial vitamin D levels and early recurrent stroke in patients presenting with acute ischemic stroke (AIS).

Material and Methods: Four hundreds and ten consecutive patients who were followed-up with AIS diagnosis were included in the study. Risk factors for ischemic stroke were questioned and biochemical and cardiac examinations of the patients were performed. Simultaneously, the National Institutes of Health Stroke Scale (NIHSS) was calculated. Patients who had recurrent ischemic stroke obtained retrospectively were from the hospital registry system in last three month. The patients were divided into two groups as those who had recurrent stroke and who did not.

Results: A total of 410 patients, 187 of which were female (45.6%) and 223 of which were male (54.4%) with a mean age of 68.12 ± 12.63 (29-99) were included in the study. Early recurrent ischemic stroke was detected in 48 (11.7%) patients in the follow-up. The systolic blood pressure and diastolic blood pressures at the time of first consultation were higher in the recurrent ischemic stroke group compared to the non-recurrent ischemic stroke group (p<0.001). Serum total vitamin D and serum parathyroid hormone levels in patients without recurrent stroke were higher as compared with those in patients with recurrent stroke (p<0.001). 25(OH) D were significantly associated with stroke recurrence (OR:1.277;CI 95%, 1.166-1.398; p<0.001).

Conclusion: Our study showed that decreased serum 25 (OH)D in patients presenting with acute ischemic stroke is associated with recurrent ischemic stroke in the early stage.

Keywords: Ischemic stroke; recurrent stroke; Vitamin D

INTRODUCTION

Stroke is one of the leading causes of mortality and morbidity throughout the world (1). Although the mortality rate has decreased with new developments in treatment, morbidity rate continues to increase (2). Two-fold increase in mortality and disability rate has been observed, especially with recurrent stroke (3). Determining the risk of recurrent stroke in the early stage may play a significant role in reducing mortality and morbidity (4). While traditional risk factors such as age, sex, hypertension, diabetes, and smoking may increase the risk of recurrent stroke, all risk factors cannot be individually held responsible for clinical recurrent stroke.

Decreased vitamin D has been reported as a new risk factor for stroke in recent years (5-7). The pathophysiological mechanisms underlying the relationship between decreased 25-hydroxyvitamin D [25(OH)D] and poor quality of life after stroke are discussed. In animal models, it has been shown that vitamin D reduces thrombosis and inflammatory mediators such as interleukin 6 (IL-6) and tumor necrosis factor alpha (TNF-alpha), preventing ischemic stroke by (8,9). In particular, decreased plasma concentrations of 25(OH) D have been found to gradually increase the risk of symptomatic ischemic stroke (10). Although this relationship has been demonstrated in controlled studies, current data are limited. Decrease in serum vitamin D levels has been shown to increase the loss of function and mortality caused by stroke (5,6). However, the relationship between vitamin D levels and the frequency of recurrence of early stage stroke is not yet clear.

The aim of our study is to examine the relationship between initial 25(OH) D levels and early stroke recurrence in patients presenting with acute ischemic stroke (AIS).

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MATERIAL and METHODS

A total of 602 patients who were diagnosed with AIS according to their neurological examination findings, computed brain tomography (CT) and/or diffusion magnetic resonance imaging (DWI-MRI) between January 2017 and March 2019, and who were followed up in the neurology clinic, were retrospectively reviewed. Patients with subacute ischemic stroke, intracerebral hemorrhage, autoimmune diseases, and patients receiving vitamin or calcium replacement therapy were excluded from the study. Local ethics committee approval was obtained for the study.

Detailed physical and neurological examinations of all patients were repeated. Age, sex, smoking, hypertension, heart disease, diabetes mellitus, and transient ischemic attack histories of the patients were questioned. Systolic blood pressure (SBP) and diastolic blood pressure (DBP) values measured at the first admission were recorded. Full blood count, renal function tests, fasting blood glucose and lipid profile including low density lipoprotein (LDL), total cholesterol and high density cholesterol (HDL) were analyzed.

Blood samples of the patients were analyzed in the first 24 hours following admission for serum 25(OH) D levels. Serum Total Vitamin D and serum parathyroid hormone (PTH) assays analyzed using automated direct competitive chemiluminescent immunoassay method by the Siemens ADVIA Centaur XP system (Siemens Healthcare United Kingdom). Venous blood samples were taken into gel biochemical tubes and centrifuged within 30 minutes at 4000 rpm for 10 minutes. Serum total Vitamin D reference range was 30-100 ng/ml and serum PTH's reference range is 19-88 pg/ml.

Stroke was assessed according to the TOAST classification and the clinical stroke syndrome classification was performed based on the Oxfordshire Community Stroke Project (OCSP) (11,12). The National Institutes of Health Stroke Scale (NIHSS) scores were calculated.

Early recurrence of ischemic stroke was defined as worsening of functional neurological status, increase in NIHSS at least 4 points or new pathological imaging findings (MRI or CT) with new focal neurological deficit after index stroke within 90 days. Patients who had recurrent stroke in last three months were obtained from the hospital registry system, retrospectively. Patients whose records could not be reached or whom died were excluded from the study. The survivals of patients were evaluated by their TR ID numbers in Republic of Turkey Ministry of Health, Directorate of Public Health, Death Notification System (https://obs.gov.tr/).

Statistical Analysis

SPSS 20.0 for Windows (SPSS Inc., Chicago, IL, USA) was used for statistical analyses were. The normally distributed continuous measurements were presented as the mean and standard deviation. The distribution of the variables was evaluated with Kolmogorov-Smirnov test. Student T and Chi-square tests were used to compare numeric and string variables, respectively. Spearman rank correlation was used for correlation analysis. For statistically significant ($p<0.05$) parameters in univariate analysis, a multiple logistic regression analysis was performed. P values less than 0.05 were considered to indicate statistical significance.

RESULTS

From the 410 of 602 AIS patients (68.1%) whose hospital recordings able to reach at the last three months, included to the study. 187 of the patients were female (45.6%) and 223 were male (54.4%) and the mean age was 68.12 ± 12.63 (29-99). Forty-five patients (7.4%) died during the follow-up. The median NIHSS score was 6.49 (2 ± 19). The mean of 25 (OH) D level was 16.8 ng /ml (4.09 ± 46.43 ng / ml) in all patients. 25(OH) D levels of 275 patients (67.1%) were determined to be below 20 pg/dl. The most common cause of cerebral stroke was cardioembolism (35.9%). While lacunar type strokes were more common in patients with recurrent AIS, strokes due to posterior circulation were more frequent in non-recurrent AIS patients (p=0.036; p=0.021). The clinical and demographic characteristics of the patients are provided in Table 1.

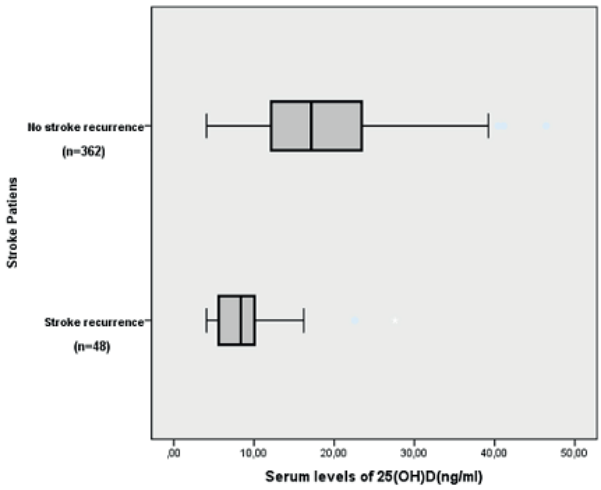

Figure 1. Distribution of 25 (OH) vitamin D levels in patients with and without stroke recurrence. Vitamin D levels were lower in the group with recurrent stroke compared to the group without recurrent stroke significantly ($p<0.001$)

Recurrent stroke was detected in 48 (11.7%) patients. SBP measured on the first day was mean ± SD 179.5±13.6 mmHg and was significantly higher in the recurrent stroke group ($p<0.001$). In addition, SBP remained significant predictors for recurrent stroke (OR:0.907;Cl 95%, 0.875-0.941; P<0.001). DBP was determined to be higher in recurrent ischemic stroke group ($p<0.001$). Serum 25(OH) D levels were significantly lower in patients with recurrent stroke than non-recurrent patients ($p<0.001$) (Figure 1). In multivariate logistic regression analysis, we calculated the odds ratio (OR) of 25(OH) D levels as compared with other risk factors. 25(OH) D were significantly associated with stroke recurrence (OR:1.277;Cl 95%, 1.166-1.398; $p<0.001$). PTH levels were significantly higher in recurrent

Table 1. Demographic data of acute ischemic stroke patients

	Without Stroke Recurrens Patients	With Stroke Recurrens Patients	p*
	n=362	n=48	
Age	68.3±12.5	66.2±13.4	0.275
Male (%)	193(53.3)	30(62.5)	0.230
Married (%)	230(63.5)	34(70.8)	0.467
Risk Factors (%)			
Hypertension	247(68.2)	32(66.7)	0.950
Diabetes mellitus	128(35.4)	14(29.2)	0.827
Transient ischemic attack	88(24.3)	26(54.2)	0.761
Hyperlipidemia	154(42.5)	17(35.4)	0.885
Atrial fibrillation	59(16.3)	7(14.6)	0.713
Tobacco	104(28.7)	14(29.2)	0.950
Pre-stroke Treatment (%)			
Antiplatelet agents	95(26.2)	18(37.5)	0.948
Antikoagülan agents	26(7.2)	5(10.4)	0.426
t-PA Treatment(%)	50(13.8)	7(14.6)	0.885
NIHSS	6.48(±4.1)	6.5(±3.6)	0.977
Stroke Etiology (%)			
Large-vessel occlusive	90(24.9)	13(27.1)	0.739
Cardioembolic	132(36.5)	15(31.3)	0.479
Small-vessel occlusive	66(18.2)	11(22.9)	0.435
Other	13(3.6)	1(2.1)	0.589
Unknown	61(16.9)	8(16.7)	0.974
Stroke Syndrome (%)			
TACS	95(26.2)	10(20.8)	0.420
PACS	107(29.6)	19(39.6)	0.127
LACS	49(13.5)	12(25)	**0.036**
POCS	111(30.7)	7(14.6)	**0.021**

*The χ2 test was used for non-continuous variables; Continuous variables were compared between the groups by the Student's t-test. P value of <.05 was considered as significant. t-PA:Tissue Plasminogen Activator; NIHHS: National Institutes of Health Stroke Scale; TACS: Total Anterior Circulation Syndrome; PACS:Partial Anterior Circulation Syndrome; LACS: Lacuner Syndrome; POCS: Posterior Circulation Syndrome

Table 2. Clinical features and laboratory findings all pateints

	Without Stroke Recurrens Patients	With Stroke Recurrens Patients	p*
	n=362	n=48	
SBP (mmHg)	160.8±12.4	179.5±13.6	**<0.001**
DBP (mmHg)	77.7±8.66	84.7±9.62	**<0.001**
25(OH) D (ng/ml)	8.56±4.30	17.91±8.30	**<0.001**
PTH (pg/ml)	87.1±99.0	141.2±100.3	**<0.001**
Glucose(mg/dL)	126.8±72.3	133.7±67.03	0.510
Creatin(mg/dL)	1.36±1.52	1.11±0.63	0.124
TC(mg/dL)	200.2±51.3	186.7±54.0	0.107
LDL-C (mg/dL)	130.8±49.4	117.4±43.9	0.055
HDL-C(mg/dL)	41.10±10.2	38.9±12.7	0.262

*Continuous variables were compared between the groups by the Student's t-test. P value of <.05 was considered as significant. 25(OH) D: 25-Hydroxyvitamin D; PTH: Parathyroid Hormone; SBP: Systolic Blood Pressure; DBP: Diastolic Blood Pressure; TC: Total Cholesterol; LDL-C: Low-Density Lipoprotein-Cholesterol; HDL-C: High-Density Lipoprotein- Cholesterol

stroke group than non-recurrent stroke group (p=0.001). There was no significant difference between recurrent stroke group than non-recurrent stroke group in other laboratory tests (p>0.05).

There was a negative correlation between serum 25(OH) D levels and SBP (r=-0.163, p=001), DBP (r=-0.115, p=0.020) measures and PTH (r=-0.230, p<0.001), and there was no significant correlation between the other parameters (Table 2).

DISCUSSION

In recent years, there have been studies investigating the possible role of vitamin D in preventing stroke, reducing post-stroke mortality and morbidity (10,13-16). Renin-angiotensin-aldosterone (RAA) system is more activated in vitamin D deficiency. This contributes to the occurrence of hypertension and formation of stroke (17). The antihypertensive effect of vitamin D is due to its suppression of the RAA system and its vasculoprotective and anti-inflammatory properties. The studies conducted on rats found that renin production was suppressed by the injection of 1.25 hydroxyvitamin D (17). Locally synthesized 1,25 dihydroxyvitamin D is affected by plasma 25(OH) D levels. This suggests that 25(OH) D may be responsible for the hormonal effects of vitamin D (18). Additionally, vitamin D receptors are found in many vascular tissues and brain cells (8). It causes atherosclerotic changes in vascular smooth muscle and foam cells in macrophages, and plays a role in the development of proatherothrombotic processes. It can prevent the formation of thrombosis by affecting the clotting mechanisms of vitamin D (for example, plasminogen activator inhibitor type 1 suppresses and activates thrombomodulin). Vitamin D deficiency is closely associated with other constituents of the metabolic syndrome, such as hypertension and diabetes, and should be considered as a risk factor for stroke.

Decreased vitamin D levels are associated with inflammation, increased calcium accumulation in the coronary arteries, and increased deterioration of endothelial function and vascular structure. First, Kilkkinen et al. determined a significant relation between 25(OH)D level and the development of fetal ischemic stroke; found that this relationship is not involved in hemorrhagic stroke (13). In their formative study including 10,170 patients, Jacobsen et al. reported that 25(OH)D levels were associated with the increase of the risk of ischemic stroke (10). Zhour et al. found that decreased vitamin D was a risk factor for the development of ischemic stroke (19). In another study, while a strong association was determined in 464 female patients between decreased 25(OH)D levels and ischemic stroke, the strongest association was reported with lacunar infarction (18). While vitamin D deficiency is considered as a risk factor in the development of AIS, the association between recurrent stroke etiology and vitamin D levels is conflicting. H. Huang et al. did not determine a significant relationship between vitamin D levels and stroke subtypes in 349 recurrent stroke patients (20). Similarly, Ford et al. suggested that vitamin D replacement therapy was ineffective in protection against stroke in patients with decreased vitamin levels (21). In our study, vitamin D levels were found to be low in all patients with AIS. In particular, decreased levels of vitamin 25(OH) D were found to be an independent risk factor that increased the frequency of recurrent stroke during follow-up. Due to the geographical conditions, low sun exposure and racial differences, the secondary vitamin D is thought to be low. Consequently, in the light of these data, the normalizating the vitamin D levels may be important in the follow-up of patients in terms of both hypertension development and prevention of ischemic stroke.

Several studies have supported that increased PTH levels correlate pro-atherosclerosis, vessel wall dysfunction, heart failure and stroke. (22-25). There are many theories about eleveted PTH levels is the leading cause of vitamin D deficiency. Firstly, 25(OH)D deficiency, which plays a key role in calcium homeostasis, causes an increase in PTH secretion as a result elevated PTH levels have been found to be associated with the risk of stroke (22). Furthermore, increased PTH levels are associated with inflammation that are increase cardiovascular risk and cerebrovasculer complication (24). Incidence of cardiovascular-associated mortallity is reduced after PTH levels are decreased by parathyroidectomy and specific drugs (24). Sato et al. observed a decrease in serum 25(OH)D levels and an increase in serum calcium and PTH levels in female subjects with ischemic stroke (25). In another study Celik et al. was claimed that the increase in PTH levels a mild risk factor for recurrent stroke. However, low levels of 25 (OH) D and elevated PTH levels together could be used as a marker in recurrent strokes (26). In our study, while a negative correlation was determined between 25(OH) D and PTH levels, a significant increase was observed in PTH levels in the recurrent stroke group compared to the non-recurrent stroke group.

Following the ischemic stroke, recurrent stroke often occurs in the first days or weeks. In the literature, the frequency of recurrent stroke is reported to be in the range of 10.0-17.7% (27-30). This wide range has been associated with patients of different ethnic groups. In parallel with the literature, we have determined the frequency of recurrent stroke as 11.7% in the 3-month follow-up in our study.

It is reported that there is a correlation between elevated systolic and diastolic blood pressure and stroke and stroke recurrence at admission (31). It has been reported that increase in SBP and DBP above 10 mmHg in ischemic stroke follow-up is associated with an increase in the risk of stoke recurrence (32). In recent meta-analyzes, it has been found that blood pressure control reduced the frequency of recurrent stroke in the long term (33,34). In our study, it has been determined that SBP and DBP were significantly higher in the recurrent stroke group, especially in the acute stage, and were significantly negatively correlated with decreased vitamin D.

Our study had certain limitations. The primary limitation was that our study was conducted in a single center. In addition, although we performed multivariate analysis, parameters such as additional diseases that could affect 25(OH) D levels (e.g. dementia) were not included in the statistical analysis. However, the fact that we did not take consider the seasonal characteristics, that we could check 25(OH) D levels in stroke patients only once and that we did not have sufficient information about 25(OH) D levels at the time of the recurrence of the stroke are the other limitations.

CONCLUSION

Our study has shown that decreased 25(OH)D level is a risk factor for early stroke recurrence. The importance of vitamin D in reducing mortality and morbidity is increasing day by day. Although vitamin D deficiency is known to be a risk factor for ischemic stroke, there is a need for experimental and clinical investigations to investigate its possible role on stroke severity and prognosis due to its neuroprotective propertie.

Competing interests: The authors declare that they have no competing interest.

Financial Disclosure: There are no financial supports.

Ethical approval: This study was approved by the Institutional Ethics Committee and conducted in compliance with the ethical principles according to the Declaration of Helsinki.

REFERENCES

1. Feigin VL, Forouzanfar MH, Krishnamurthi R, et al. Global and regional burden of stroke during 1990-2010: findings from the Global Burden of Disease Study 2010. Lancet 2014;383:245-54.
2. Wang W, Jiang B, Sun H, et al. Prevalence, Incidence, and Mortality of Stroke in China: Results from a Nationwide Population-Based Survey of 480 687 Adults. Circulation 2017;135:759-71.
3. Zhang Q, Ding H, Yan J, et al. Plasma tissue kallikrein level is negatively associated with incident and recurrent stroke: a multicenter caseecontrol study in China. Ann Neurol 2011;70:265-73.
4. Wang A, Wu L, Wang X, et al. Effect of recurrent stroke on poor functional outcome in transient ischemic attack or minor stroke. Int J Stroke 2016;11:80-2.
5. Kojima G, Bell C, Abbott RD, et al. Low dietary vitamin D predicts 34-year incident stroke: The Honolulu Heart Program. Stroke 2012;43:2163-7.
6. Pilz, S, Dobnig H, Fischer JE, et al. Low vitamin d levels predict stroke in patients referred to coronary angiography. Stroke 2008;39:2611-3.
7. Buell JS, Dawson-Hughes B, Scott TM, et al. 25-Hydroxyvitamin D, dementia, and cerebrovascular pathology in elders receiving home services. Neurology 2010;74:18-26.
8. Bouillon R, Carmeliet G, Verlinden L, et al. Vitamin D and human health: Lessons from vitamin D receptor null mice. Endocr Rev 2008;29:726-76.
9. Guillot X, Semerano L, Saidenberg-Kermanac'h N, et al. Vitamin D and inflammation. Jt. Bone Spine 2010;77:552-7.
10. Brøndum-Jacobsen P, Nordestgaard BG, Schnohr P, et al. 25- Hydroxyvitamin D and symptomatic ischemic stroke: an original study and meta-analysis. Ann Neurol 2013;73:38-47.
11. Adams Jr HP, Bendixen BH, Kappelle LJ, et al. Classification of subtype of acute ischemic stroke. Definitions for use in a multicenter clinical trial. TOAST. Trial of Org 10172 in acute stroke treatment. Stroke 1993;24:35-41.
12. Bamford J, Sandercock P, Dennis M, et al. Classification and natural history of clinically identifiable subtypes of cerebral infarction. Lancet 1991;337:1521-6.
13. Kilkkinen A, Knekt P, Aro A, et al. Vitamin D status and the risk of cardiovascular disease death. Am J Epidemiol 2009;170:1032-9.
14. Schierbeck LL, Rejnmark L, Tofteng CL, et al. Vitamin D deficiency in postmenopausal, healthy women predicts increased cardiovascular events: A 16-year follow-up study. Eur. J. Endocrinol 2012;167:553-60.
15. Alfieri DF, Lehmann MF, Oliveira SR, et al. Vitamin D deficiency is associated with acute ischemic stroke, C-reactive protein, and short-term outcome. Metab Brain Dis 2017;32:493-502.
16. Leung RY, Han Y, Sing, CW, et al. Serum 25-hydroxyvitamin D and the risk of stroke in Hong Kong Chinese. Thromb Haemost 2017;117:158-63.
17. Li YC, Qiao G, Uskokovic M, et al. Vitamin D: a negative endocrine regulator of the renin-angiotensin system and blood pressure. J Steroid Biochem Mol Biol 2004;89-90:387-92.
18. Sun Q, Pan A, Hu FB, et al. 25-Hydroxyvitamin D levels and the risk of stroke: a prospective study and meta-analysis. Stroke 2012;43:1470-7.
19. Zhou R, Wang M, Huang H, et al. Lower Vitamin D Status Is Associated with an Increased Risk of Ischemic Stroke: A Systematic Review and Meta-Analysis. Nutrients 2018;10:277.
20. Huang H, Zheng T, Wang S, et al. Serum 25-hydroxyvitamin D predicts early recurrent stroke in ischemic stroke patients. Nutr Metab Cardiovasc Dis 2016;26:908-14.
21. Ford JA, MacLennan GS, Avenell A, et al. Cardiovascular disease and vitamin D supplementation: Trial analysis, systematic review, and meta-analysis. Am J Clin Nutr 2014;100:746-55.
22. Welles CC, Whooley MA, Karumanchi SA, et al. Vitamin D deficiency and cardiovascular events in patients with coronary heart disease: data from the Heart and Soul study. Am J Epidemiol 2014;179:1279-87.
23. Fitzpatrick LA, Bilezikian JP, Silverberg SJ. Parathyroid hormone and the cardiovascular system. Curr Osteoporos Rep 2008;6:77-83.
24. Hagström E, Hellman P, Larsson TE, et al. Plasma parathyroid hormone and the risk of cardiovascular mortality in the community. Circulation 2009 Jun 2;119:2765-71.

25. Sato Y, Kaji M, Metoki N, et al. "Does compensatory hyperparathyroidism predispose to ischemic stroke?" Neurology 2003;60:626-9
26. Celik G, Dogan A, Dener S, et al. Parathyroid Hormone Levels in the Prediction of Ischemic Stroke Risk. Dis Markers 2017;2017:4343171
27. Howard G, Kissela BM, Kleindorfer DO, et al. Differences in the role of black race and stroke risk factors for first vs recurrent stroke. Neurology 2016;86:637-42.
28. Aarnio K, Haapaniemi E, Melkas S, et al. Long-term mortality after first-ever and recurrent stroke in young adults. Stroke 2014;45:2670-6.
29. Giang KW, Björck L, Ståhl CH, et al. Trends in risk of recurrence after the first ischemic stroke in adults younger than 55 years of age in Sweden. Int J Stroke 2016;11:52-61.
30. Wang Y, Xu J, Zhao X, et al. Association of hypertension with stroke recurrence depends on ischemic stroke subtype. Stroke 2013;44:1232-7.
31. Hong KS. Blood Pressure Management for Stroke Prevention and in Acute Stroke. J Stroke 2017;19:152-65.
32. Chang Y, Kim J, Kim YJ, et al. Inter-arm Blood Pressure Difference is Associated with Recurrent Stroke in Non-cardioembolic Stroke Patients. Sci Rep 2019;9:12758.
33. Lisheng L, Zengwu W, Yuging Z, et al. Blood pressure reduction for the secondary prevention of stroke: a Chinese trial and a systematic review of the litera¬ture. Hypertens Res 2009;32:1032-40.
34. Katsanos AH, Filippatou A, Manios E, et al. Blood pressure reduction and secondary stroke prevention: a systematic review and metaregression analysis of randomized clinical trials. Hypertension 2017;69:171-9.