Decreased Heart Rate Variability is Associated with Increased Transcranial Doppler Velocities in Children with Sickle Cell Disease

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Objective: To explore the relationship between 24-hour blood pressure (BP) variability, heart rate (HR) variability, and transcranial Doppler velocity (TCDV) in a cohort of pediatric sickle cell disease (SCD) patients.

Design, Setting, and Participants: This is a retrospective study of 11 children aged 8–18 years with SCD who previously underwent 24-hour ambulatory BP monitoring and TCDV measurements.

Interventions: Medical records were reviewed for TCDV and 24-hour ABP data. TCDV in the right and left middle cerebral artery were examined, and the highest velocity was recorded. HR and BP standard deviations were used as markers of variability. The relationships between daytime, nighttime, and 24-hour blood pressures and heart rate variability were determined.

Results: Mean age, body mass index and hemoglobin levels were 11.2 ± 3.0 years, 18.7 ± 3.4 kg/m^2, and 9.1 ± 1.7 g/dL, respectively. Median transcranial Doppler velocity was 136cm/s (125–142). Decreased day, night, and 24-hour heart rate variability were significantly associated with increased transcranial Doppler velocity ($R=-.69$, $P=.02$; $R=-.82$ $P=.002$; $R=-.66$, $P=.03$, respectively). BP variability did not correlate with TCDV. Nighttime BP indexes were higher than daytime.

Conclusions: In this small cohort, decreased heart rate variability assessed by the standard deviation of HR was associated with increased transcranial Doppler velocities in children with SCD. No correlation between measurements of BP variability and TCDV was found. Our study provides new information on heart rate and blood pressure variability and TCDV; a surrogate marker of stroke risk in sickle cell disease. Larger multicenter studies are needed to confirm our findings. (*Ethn Dis*. 2014;24[4]:451–455)

Key Words: Sickle Cell Anemia, Transcranial Doppler Ultrasonography, Ambulatory Blood Pressure Monitoring, Blood Pressure Variability, Pediatric

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Introduction

Sickle cell disease (SCD) is associated with multiple comorbidities and remains one of the world's most common hereditary disorders.[1] Individuals with SCD suffer repeated vaso-occlusive events characterized by ischemia-reperfusion injury and inflammation.[2,3] These chronic vascular insults lead to numerous target-organ complications such as avascular necrosis of bones, retinal infarction, stroke, acute chest syndrome, pulmonary hypertension, and skin ulceration. A seminal feature that sets sickle cell disease apart from other chronic hemolytic syndromes and that predicts disease severity is a chronic, intense inflammatory state. Sickle cell disease is a chronic inflammatory condition with an ongoing insult to the vascular tree and increased prevalence of microalbuminuria, oxidative stress and iron overload, abnormal nitric oxide, hypoxia and reoxygenation injury, inflammatory cytokines, leukocytosis and increased inflammatory markers such as CRPs.[4–6]

Stroke is a devastating SCD morbidity and affects as many as 11% of children who inherit sickle hemoglobin from both parents (HbSS).[7–11] The pathophysiology of stroke is multifactorial with vasculopathy due to ongoing inflammation and a state of nitric oxide (NO) resistance recently taking a central role. Wood et al[12] described sickle cell disease as a steady state of increased plasma cell-free hemoglobin and overproduction of reactive oxygen species (ROS) that scavenge endothelium-derived NO and metabolize its precursor arginine, impairing NO homeostasis. Human physiological and transgenic animal studies provide experimental evidence of cardiovascular and pulmonary resistance to NO and reduced NO bioavailability that is associated with vasoconstriction, decreased blood flow, platelet activation, increased endothelin-1 expression, and end-organ injury.

Transcranial Doppler (TCD) ultrasonography is a cost-effective, noninvasive tool used to identify vessel narrowing or occlusion within the large cerebral vessels to assess risk of stroke in children with SCD.[13] Measured velocities greater than 200 cm/s in the large cerebral vessels are considered abnormal and associated with a higher risk of stroke. Healthy children have an average transcranial Doppler velocity (TCDV) of 79 ± 13 cm/s, while SCD children without overt stroke average 133 ± 19 cm/s in the middle cerebral artery (MCA).[14]

24-hour ambulatory blood pressure monitoring (ABPM) provides superior assessment of blood pressure (BP) measurements and patterns as well as heart rate (HR). Studies have shown ABPM to better predict cardiovascular mortality than clinic blood pressure measurements;[15] ABPM is valuable in determining BP and HR variability using the standard deviation of 24-hour ABP and HR readings.[16] Blood pressure varies considerably throughout daytime hours and to a lesser extent during the night to accommodate differing ambulatory and environmental changes in normal persons.[17]

Heart rate variability occurs in response to specific physical demands. Sympathetic and parasympathetic autonomic nervous system influences dynam-

ically regulate the degree of HR variability;[18] HR variability is known to decrease with stress and increase during times of rest.[19] Reduced HR variability indicates an autonomic imbalance, possibly caused by weak parasympathetic control,[20] which endorses sympathetic dominance and consequently mortality.[21] Previous studies have shown an association between reduced HR variability and increased all-cause mortality and adverse cardiovascular events.[21–23]

Incidence and severity of target organ damage has been shown to correlate with increased 24-hour BP variability in the hypertensive adult population in several previous studies.[24–28] Evidence also suggests that variability and instability in blood pressure increases the risk of vascular events, especially stroke.[25,29]

The relation of BP and HR variability to risk of stroke in the pediatric SCD population has not been previously defined. In our study, we examine the relationship between HR variability, BP variability, and TCDV in pediatric sickle cell patients. We hypothesize that decreased HR variability and increased BP variability are associated with increased TCDV, and therefore increased cardiovascular disease risk and stroke.

METHODS

Setting and Study Design

Medical records of eleven patients aged 8–18 years old with SCD who previously underwent 24-hour blood pressure monitoring were used to examine 24-hour BP variability and HR variability and their relationship to TCDV values. Demographic and biochemical data were collected. Approval for this study was granted by the Institutional Review Board at the Medical University of South Carolina (MUSC).

We hypothesize that decreased HR variability and increased BP variability are associated with increased TCDV, and therefore increased cardiovascular disease risk and stroke.

Children were included if they had Hb electrophoresis-confirmed SCD (HbSS, HbSβ^0 thalassemia, and HbSC) and were aged 8–18 years at the time of the study. Participants were ineligible for 24-hr ABP monitoring if they were in pain at time of clinic visit, had vaso-occlusive crisis (VOC), acute chest syndrome (ACS) in the last 3 weeks (to ensure that pain is not contributing to BP abnormalities), had other concomitant primary kidney disease, or secondary hypertension, were receiving angiotensin-converting enzyme inhibitors (ACEi) or angiotensin II receptor blockers (ARBs).

Patients excluded secondary to pain, VOC, or ACS were eligible if they presented subsequently for follow up and were pain-free during the study enrollment period.

Chronic transfusion treatment (defined as receiving packed erythrocyte transfusion or erythrocytapheresis approximately every 4 weeks with the goal of maintaining a pre-transfusion hemoglobin S concentration between 30–50% for at least 2 years) was recorded as this subgroup of patients generally have a more severe disease (eg, an abnormal TCD measurement, history of stroke).

Assessment of ABPM

The patients participated in a previous MUSC ABP monitoring study where 24-hour ABP was recorded utilizing the Spacelabs 90217 ambulatory monitor (Spacelabs Medical, Issaquah, WA), fitted and programmed as previously described.[30,31] Mean and standard deviation of daytime, nighttime, and 24-hr systolic and diastolic BP values were extracted from the study records. Ambulatory systolic and diastolic BP indexes (mean daytime and night time ambulatory BP divided by the 95th percentile for pediatric ambulatory BP normative data) were calculated; HR and BP standard deviations were used as markers of variability.[16]

Participants also had an ABP study visit while not having any concurrent acute medical illnesses that included a history and a physical examination. Body weight and standing height were measured using a digital scale and a wall-mounted stadiometer to the nearest 1 kg and .5 cm, respectively. BMI was calculated by dividing the weight in kilograms by the square of the height in meters.

Ambulatory blood pressure monitoring and TCD measurements were done in the outpatient setting within no more than one year. The ABPM machines were programmed to obtain a measurement every 20 minutes during the daytime and every 30 minutes during the sleep period. A successful ABPM was defined as $>$ 80% successful readings with at least one successful measurement every hour.

Assessment of TCDV

Velocities of TCD were extracted from the medical records of the study population. The TCD studies were performed using the Spencer Technologies ST3 ultrasound apparatus with a transducer operating at 2 MHz. The right and left middle cerebral artery, anterior cerebral artery, intracavernous internal carotid artery, and posterior cerebral artery were analyzed using a standard transtemporal approach. Only MCA velocities were reported for the purposes of our study because the MCA has typically high blood flow velocities and is a common site of infarct. The MCA was identified with flow direction toward the probe and the signal was optimized. Samples of the MCA were isonated along a depth of 30mm–56mm, depending on the size of the child's head, at 2mm increments up to the MCA/ACA bifurcation. Velocities described in this

Table 1. Demographic and bioclinical characteristics of the study population[a]

Variable	All SCD Patients (*n*=11)
Age, years	11.2 ± 3.0
Sex, *n* (%)	
Male	7 (64%)
Female	4 (36%)
BMI, kg/m^2	18.7 ± 3.4
Receiving chronic transfusion therapy	5 (45%)
TCDV, cm/s	136 (125, 142)
Hb level, g/dL	9.1 ± 1.7

SCD, Sickle cell disease; TCDM, transcranial Doppler velocity.
[a] Continuous, normally distributed variables are presented as mean ± SD and categorical variables presented as *n* (%). Continuous, not normally distributed variables, are presented as medians and interquartile ranges.

article are time-averaged means of Vmax in either the right or left MCA, depending on which was the greatest value.

Methods of Statistical Analysis

Categorical variables are presented as relative frequencies. Continuous variables, not normally distributed, are presented as medians and interquartile ranges. Normally distributed continuous variables are presented as means ± standard deviations. Pearson correlation was performed between ABP parameters and TCDV. A multivariable linear regression model with TCD as outcome variable and night HRV as variable of interest was built; variables of clinical significance (age, BMI and Hb level) were also included in the model.

Statistical and database software used included STATA 9.2 (Stata Corporation, College Station, TX) and Microsoft Office Excel 2007 (Microsoft Corporation, Redmond, WA), respectively. Statistical significance for all analyses was set at two-sided $P<.05$.

Table 2. 24-hr indexed ABP and HR measurements[a]

Variable	All SCD Patients (*n*=11)
24-hr systolic index	.97 (.91, 1.05)
24-hr diastolic index	.93 (.85, 1.00)
Day systolic index	.95 (.93, 1.02)
Day diastolic index	.91 (.85, 1.00)
Night systolic index	.99 (.94, 1.10)
Night diastolic index	1.00 (.86, 1.10)
24-hr heart rate	81 (78, 96)

[a] Ambulatory systolic and diastolic BP indexes (mean daytime and night time ambulatory BP divided by the 95th percentile for pediatric ambulatory BP normative data), presented as median (interquartile range).

RESULTS

Complete data and TCD studies were available for 11 children. All were African-American, 64% were male, mean BMI was 18.7 ± 3.4 kg/m^2, five of the children (45%) were receiving chronic transfusion therapy at the time of the study. The TCDV measurements were all taken from either the right or left MCA, depending on which reading was highest. Median TCDV was 136cm/s (125, 142 cm/s). (Table 1)

Indexed BP variables are presented in Table 2. Nighttime BP indexes were higher than daytime. This is consistent with our finding in the full cohort of 39 children who underwent ABPM.[32] Night-time diastolic index had a median of 1.00, indicating that as a group, children had a night-time diastolic BP that met the threshold for being hypertensive.

Heart rate variability was inversely and significantly associated with TCDV. The strongest inverse correlation was found between night-time HR variability and TCDV ($R=-.82$, $P=.002$), Figure 1. A statistically significant inverse correlation also existed between daytime HR variability and TCDV ($R=-.69$ $P=.02$) and between 24-hr HR variability and TCDV ($R=-.66$, $P=.03$), Table 3. In a multivariable linear regression model of TCD determinants; after adjusting for age, BMI, and Hb levels, night HRV was significantly associated with TCD velocity, Table 4. No significant correlation was

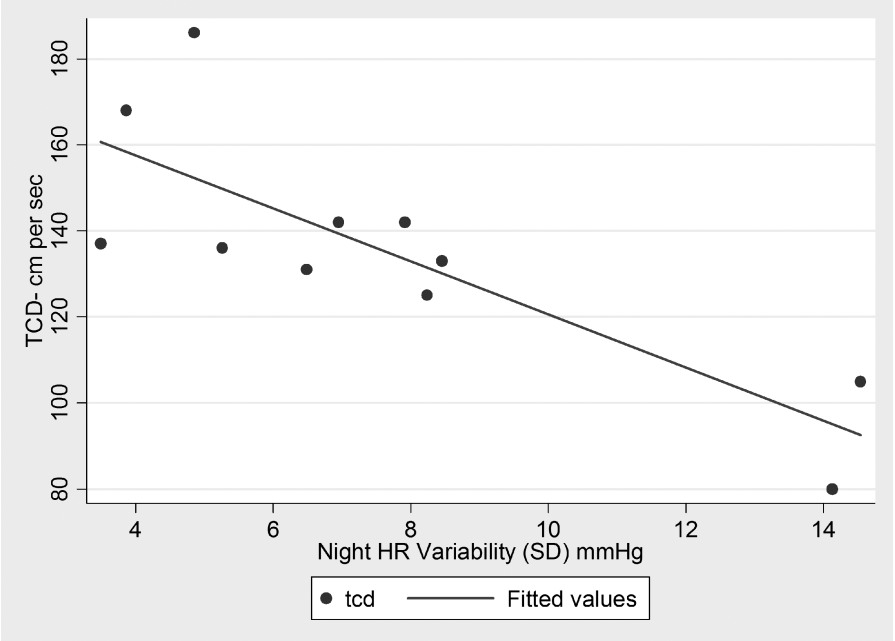

Fig 1. Night-time heart rate/blood pressure variability and transcranical Doppler velocity

Table 3. Bivariate correlation between TCD velocities and HR variability[a]

	Day HR SD	Night HR SD	24-hr HR SD
TCDV	−.69 (.02)	−.82 (.002)	−.66 (.03)

TCDV, transcranial Doppler velocity; HR, heart rate.
[a] Data are Pearson correlation coefficient (*P*).

In our study, we show for the first time that reduced HR variability assessed by standard deviation is significantly associated with increased TCDV in children with SCD. Day-time, night-time, and 24-h HR variability were all inversely correlated with TCDV.

found between BP variability and TCDV (data not shown).

DISCUSSION

The effect of HR and BP variability on risk of stroke in children with sickle cell disease is yet undefined. In our study, we show for the first time that reduced HR variability assessed by standard deviation is significantly associated with increased TCDV in children with SCD. Day-time, night-time, and 24-h HR variability were all inversely correlated with TCDV. Consistent data exist within various alternate populations; Binici et al reported a strong correlation between reduced night-time HR variability and stroke risk among 653 healthy adult participants with no history of stroke or cardiovascular disease.[20] Pringle et al[33] demonstrated night-time systolic BP variability as an independent risk factor for stroke in an elderly hypertensive population, whereas Tatasciore et al[34] found a positive correlation between daytime systolic BP variability and target organ damage in a younger hypertensive population.

Decreased HR variability is indicative of an autonomic imbalance and has been reported in multiple disease states such as diabetes, hypertensive cardiac hypertrophy, and atherosclerosis.[19] In studies examining cardiovascular characteristics following stroke, reduced HR variability has also been shown.[35,36] In our study, the relationship between TCD velocity and night-time HRV continued to be significant after adjusting for age, BMI and Hb levels.

Increased BP variability is thought to induce organ damage by inflicting trauma on blood vessels walls as BP fluctuates.[31] Several previous studies have consistently suggested a relationship between increased variability and target organ damage and risk of stroke in hypertensive populations. Our study did not support any correlation between BP variability and TCDV; this is possibly related to our small sample size or different characteristics of the cardiovascular tree of children with sickle cell disease. Additionally, previous work suggests that if the BP values are obtained at intervals greater than 15 minutes, then the accuracy of SD as an assessment of BP variability is low.[37]

Identifying additional, modifiable and easily measurable risk factors of stroke is clinically important to reduce and possibly prevent this devastating morbidity of SCD. Children with SCD are at risk for developing stroke, especially if the child has increased transcranial Doppler velocities in the large cerebral vessels. Due to the inverse relationship between blood flow velocity and vessel diameter, a higher TCDV in cerebral vessels may be indicative of vessel stenosis.[10] Though none of our study participants showed abnormal TCD velocities (>200cm/s), all 11 velocities were above the average of 79 cm/s found in healthy children and consistent with the average TCDV of children who developed strokes.[9]

Our study was limited by its retrospective design and small sample size. Standard deviations were utilized to assess variability; other time-depended measures are more sensitive. Heart rate variability can be influenced by environmental factors, physical activity, smoking, and stress. To our knowledge, none of the participants was a smoker at time of the study, all ABPM studies were started in clinic and children carried the machine to school the next day. On the other hand, it explores a potentially very important relationship between HR variability and TCDV. Also, our study was limited to patients 8–18 years old, future prospective studies examining this relationship in patients younger than 8 years of age are needed.

Decreased HRV is associated with increased TCD velocities in children with SCD. Our study provides new

Table 4. Multivariable regression model of TCD determinants

Variable	Regression Coefficients	*P*	95% Confidence Interval
Night HRV (SD)	−7.5	.02	−13.1, −1.9
Age	1.2	.6	−3.9, 6.3
BMI	.2	.93	−4.7, 5.1
Hemoglobin	4.4	.4	−6.9, 15.7

HRV, heart rate variability; TCD, transcranial Doppler.

information on HR and BP variability and TCDV, a surrogate marker of stroke risk in SCD. Larger multicenter studies are needed to confirm our findings using a more sensitive measure of HR and BP variability.

REFERENCES

1. Sankaran VG. Targeted therapeutic strategies for fetal hemoglobin induction. *Hematology Am Soc Hematol Educ Program*. 2011;2011: 459–465.
2. Kaul DK, Hebbel RP. Hypoxia/reoxygenation causes inflammatory response in transgenic sickle mice but not in normal mice. *J Clin Invest*. 2000;106:411–420.
3. Osarogiagbon UR, Choong S, Belcher JD, et al. Reperfusion injury pathophysiology in sickle transgenic mice. *Blood*. 2000;96: 314–320.
4. Jison ML, Munson PJ, Barb JJ, et al. Blood mononuclear cell gene expression profiles characterize the oxidant, hemolytic, and inflammatory stress of sickle cell disease. *Blood*. 2004;104:270–280.
5. Platt OS. Sickle cell anemia as an inflammatory disease. *J Clin Invest*. 2000;106:337–338.
6. Aslan M, Freeman BA. Oxidant-mediated impairment of nitric oxide signaling in sickle cell disease–mechanisms and consequences. *Cell Mol Biol (Noisy-le-grand)*. 2004;50:95–105.
7. Ohene-Frempong K, Weiner SJ, Sleeper LA, et al. Cerebrovascular accidents in sickle cell disease: rates and risk factors. *Blood*. 1998; 91(1):288–294.
8. Lee MT, Piomelli S, Granger S, et al. Stroke Prevention Trial in Sickle Cell Anemia (STOP): extended follow-up and final results. *Blood*. 2006;108(3):847–852.
9. Adams R, McKie V, Hsu L, et al. Prevention of a first stroke by transfusion in children with abnormal results of transcranial Doppler ultrasonography. *N Engl J Med*. 1998;339: 5–11.
10. Hokazono M, Silva GS, Silva EM, Braga JA. Results from transcranial Doppler examination on children and adolescents with sickle cell disease and correlation between the time-averaged maximum mean velocity and hematological characteristics: a cross-sectional analytical study. *Sao Paulo Med J*. 2011; 129(3):134–138.
11. Buchanan G, Vichinsky E, Krishnamurti L, Shenoy S. Severe sickle cell disease–pathophysiology and therapy. *Biol Blood Marrow Transplant*. 2010;16(1Suppl):S64–S67.
12. Wood KC1, Hsu LL, Gladwin MT. Sickle cell disease vasculopathy: a state of nitric oxide resistance. *Free Radic Biol Med*. 2008;44(8): 1506–1528.
13. Kassab MY, Majid A, Farooq MU, et al. Transcranial Doppler: an introduction for primary care physicians. *J Am Board Fam Med*. 2007;20(1):65–71.
14. Adams RJ. Big strokes in small persons. *Arch Neurol*. 2007;64(11):1567–1574.
15. Madin K, Iqbal P. Twenty four hour ambulatory blood pressure monitoring: a new tool for determining cardiovascular prognosis. *Postgrad Med J*. 2006;82:548–551.
16. Urbina E, Alpert B, Flynn J, et al. Ambulatory blood pressure monitoring in children and adolescents: recommendations for standard assessment. *Hypertension*. 2008;52:433–451.
17. Mohan JS, Marshall JM, Reid HL, Serjeant GR. Daily variability in resting levels of cardiovascular variables in normal subjects and those with sickle cell disease. *Clin Auton Res*. 1995;5:129–134.
18. Papaioannou VE. Heart rate variability, baroreflex function and heart rate turbulence: possible origin and implications. *Hellenic J Cardiol*. 2007;48(5):278–289.
19. Dekker JM, Crow RS, Folsom AR, et al. Low heart rate variability in a 2-minute rhythm strip predicts risk of coronary heart disease and mortality from several causes: the ARIC Study. *Circulation*. 2000;102:1239–1244.
20. Binici Z, Mouridsen MR, Køber L, Sajadieh A. Decreased nighttime heart rate variability is associated with increased stroke risk. *Stroke*. 2011;42(11):3196–3201.
21. Sajadieh A, Rasmussen V, Hein HO, Hansen JF. Familial Predisposition to Premature Heart Attack and Reduced Heart Rate Variability. *Am J Cardiol*. 2003;92:234–236.
22. Tsuji H, Venditti FJ Jr, Manders ES, et al. Reduced heart rate variability and mortality risk in an elderly cohort. The Framingham Heart Study. *Circulation*. 1994;90(2):878–883.
23. Tsuji H, Larson MG, Venditti FJ Jr, et al. Impact of reduced heart rate variability on risk for cardiac events. The Framingham Heart Study. *Circulation*. 1996;94:2850–2855.
24. Muller JE. Circadian variation in cardiovascular events. *Am J Hypertens*. 1999;12(2 Pt 2):35S–42S.
25. Rothwell PM. Does Blood Pressure Variability Modulate Cardiovascular Risk? *Curr Hypertens Rep*. 2011;13:177–186.
26. Mancia G, Parati G. The role of blood pressure variability in end-organ damage. *J Hypertens Suppl*. 2003;21(6):S17–23.
27. Frattola A, Parati G, Cuspidi C, Albini F, Mancia G. Prognostic value of 24-hour blood pressure variability. *J Hypertens*. 1993;11(10): 1133–1137.
28. Parati G, Pomidossi G, Albini F, Malaspina D, Mancia G. Relationship of 24-hour blood pressure mean and variability to severity of target-organ damage in hypertension. *J Hypertens*. 1987;5:93–98.
29. Geeganage C, Tracy M, England T, et al. Relationship between baseline blood pressure parameters (including mean pressure, pulse pressure, and variability) and early outcome after stroke: data from the Tinzaparin in Acute Ischaemic Stroke Trial (TAIST). *Stroke*. 2011;42(2):491–493.
30. Shatat IF, Freeman KD, Vuguin PM, Dimartino-Nardi JR, Flynn JT. Relationship between adiponectin and ambulatory blood pressure in obese adolescents. *Pediatr Res*. 2009;65:691–695.
31. Shatat IF, Flynn JT. Relationships between renin, aldosterone, and 24-hour ambulatory blood pressure in obese adolescents. *Pediatr Res*. 2011;69:336–340.
32. Shatat IF, Jakson SM, Blue AE, et al. Masked hypertension is prevalent in children with sickle cell disease: a Midwest Pediatric Nephrology Consortium study. *Pediatr Nephrol*. 2013;28(1):115–20.
33. Pringle E, Phillips C, Thijs L, et al. Systolic blood pressure variability as a risk factor for stroke and cardiovascular mortality in the elderly hypertensive population. *J Hypertens*. 2003;21(12):2251–2257.
34. Tatasciore A, Renda G, Zimarino M, et al. Awake systolic blood pressure variability correlates with target-organ damage in hypertensive subjects. *Hypertension*. 2007;50(2): 325–332.
35. Negrusz-Kawecka M, Kobusiak-Prokopowicz M. Studies of arrhythmia incidence and heart rate variability in patients suffering from cerebral stroke. *Pol Arch Med Wewn*. 1998; 100:515–525.
36. Gujjar AR, Sathyaprabha TN, Nagaraja D, et al. Heart rate variability and outcome in acute severe stroke: Role of power spectral analysis. *Neurocrit Care*. 2004;1:347–353.
37. Staessen JA, Fagard R, Thijs L, Amery A. A consensus view on the technique of ambulatory blood pressure monitoring. The Fourth International Consensus Conference on 24-Hour Ambulatory Blood Pressure Monitoring. *Hypertension*. 1995;26:912–918.