Association Between Blood Pressure Variability and Inflammatory Marker in Hypertensive Patients

Kwang-II Kim, MD*†‡; Jae-Hee Lee, RN‡; Hyuk-Jae Chang, MD*†‡; Young-Seok Cho, MD*†‡; Tae-Jin Youn, MD*†‡; Woo-Young Chung, MD*†‡; In-Ho Chae, MD*†‡; Dong-Ju Choi, MD*†‡; Kyoung Un Park, MD*†‡; Cheol-Ho Kim, MD, PhD*†‡

**Department of Internal Medicine, †Seoul National University College of Medicine, 28 Yongdong-dong, Chongno-gu, Seoul 110-744, Korea. E-mail: cheolkim@snu.ac.kr

Key Words: Ambulatory blood pressure; Blood pressure variability; Inflammation

Background  Blood pressure (BP) variability has been reported to be associated with hypertensive target organ damage and cardiovascular events. However, the exact mechanism linking BP variability and organ damage is uncertain. This study was designed to investigate the association between BP variability and inflammatory marker in hypertensive patients.

Methods and Results  Fifty-two hypertensive patients (28 men, 55.9±1.5 years) completed 24-h ambulatory BP monitoring. Inflammatory markers were evaluated by measuring plasma levels of interleukin (IL)-6, tumor necrosis factor (TNF)-α by enzyme-linked immunosorbent assay and high sensitive C-reactive protein (hs-CRP) by particle-enhanced light-scattering immunoassay. BP variability was obtained by calculating within-subject standard deviation (SD) and coefficient of variation of BP. Subjects were grouped into tertiles according to IL-6, TNF-α, and hs-CRP levels. A significant association between ambulatory BP and TNF-α level was identified (P for trend=0.011). In contrast, no association was observed between BP and IL-6 level; however, BP variability index was linked to IL-6 level (P for trend=0.046). The association between inflammatory marker and pattern of diurnal variation was investigated. The hs-CRP concentration was significantly higher in the riser group compared with the dipper group. However, IL-6 and TNF-α levels did not differ among the different diurnal variation groups. Correlation analysis showed varying associations between IL-6 and TNF-α. TNF-α level correlated with the BP index; however, IL-6 level correlated with the BP variability index. Multiple linear regression models revealed that the SD of daytime systolic BP (β=0.065, p=0.001) and age (β=0.024, p=0.016) were all positively and significantly related to IL-6. In contrast, only daytime diastolic BP (β=0.029, p=0.002) was independently related to TNF-α.

Conclusion  Inflammatory markers are associated with BP variability in hypertensive patients. This finding implies that inflammation may be a mediator for the link between BP variability and target organ damage. (Circ J 2008; 72: 293–298)

So, the identification of increased BP variability by ambulatory monitoring may be one way of detecting the high-risk subject among hypertensive patients. However, the exact mechanisms underlying the link between BP variability and cardiovascular risk are, as yet, unclear. Various mechanisms may be involved in the association between BP variability and cardiovascular disease. In addition to augmented mechanical stress on the cardiovascular system, increased variability of blood flow by augmented BP variability increases shear stress on endothelial cells. Shear stress-induced platelet activation and subsequent hypercoagulability may lead to cardiovascular events. Neurohumoral activation, which is increased in those with increased BP variability, may also increase the risk for cardiovascular disease.

One of the mechanisms explaining the relationship between BP variability and target organ damage is inflammatory response. There is some experimental evidence suggesting that elevated BP and BP variability may promote endothelial expression of cytokines and stimulate inflammation. However, the exact association between BP variability and inflammation in hypertensive subjects has not yet been evaluated. We designed this study to investigate the link between BP variability and inflammation in hypertensive patients.
Table 1  Clinical, Laboratory, and Hemodynamic Characteristics According to the IL-6 Tertile

<table>
<thead>
<tr>
<th>IL-6 tertile</th>
<th>≤0.27</th>
<th>9.27–14.87</th>
<th>&gt;14.87</th>
<th>P for trend</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>50.59±2.83</td>
<td>56.13±2.55</td>
<td>60.13±2.25*</td>
<td>0.038</td>
</tr>
<tr>
<td>Male gender</td>
<td>64.7%</td>
<td>61.1%</td>
<td>35.3%</td>
<td>0.170</td>
</tr>
<tr>
<td>Smoking</td>
<td>17.6%</td>
<td>18.8%</td>
<td>5.9%</td>
<td>0.492</td>
</tr>
<tr>
<td>LVH</td>
<td>11.8%</td>
<td>31.3%</td>
<td>11.8%</td>
<td>0.247</td>
</tr>
<tr>
<td>Albuminuria</td>
<td>23.5%</td>
<td>12.5%</td>
<td>5.9%</td>
<td>0.326</td>
</tr>
<tr>
<td>WBC (10^3/μl)</td>
<td>5.59±0.30</td>
<td>5.81±0.36</td>
<td>6.31±0.50</td>
<td>0.411</td>
</tr>
<tr>
<td>Hemoglobin (g/dl)</td>
<td>14.53±0.29</td>
<td>14.75±0.34</td>
<td>14.06±0.47</td>
<td>0.423</td>
</tr>
<tr>
<td>Platelet (10^3 /μl)</td>
<td>233.18±8.17</td>
<td>231.30±7.56</td>
<td>227.56±12.15</td>
<td>0.984</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>26.07±0.67</td>
<td>24.54±0.69</td>
<td>25.50±0.69</td>
<td>0.283</td>
</tr>
<tr>
<td>Glucose (mg/dl)</td>
<td>98.47±2.67</td>
<td>100.94±2.20</td>
<td>103.67±3.68</td>
<td>0.450</td>
</tr>
<tr>
<td>Total cholesterol (mg/dl)</td>
<td>213.12±5.89</td>
<td>204.82±8.54</td>
<td>224.63±8.08</td>
<td>0.192</td>
</tr>
<tr>
<td>TG (mg/dl)</td>
<td>141.29±17.95</td>
<td>115.00±9.76</td>
<td>113.40±8.97</td>
<td>0.255</td>
</tr>
<tr>
<td>HDL-C (mg/dl)</td>
<td>59.88±4.18</td>
<td>62.53±2.75</td>
<td>64.80±2.66</td>
<td>0.384</td>
</tr>
<tr>
<td>LDL-C (mg/dl)</td>
<td>124.98±5.58</td>
<td>117.67±8.76</td>
<td>138.52±8.79</td>
<td>0.173</td>
</tr>
<tr>
<td>Office SBP (mmHg)</td>
<td>162.82±4.61</td>
<td>160.88±3.89</td>
<td>156.47±2.82</td>
<td>0.488</td>
</tr>
<tr>
<td>Office DBP (mmHg)</td>
<td>97.52±2.37</td>
<td>96.50±2.88</td>
<td>92.24±2.41</td>
<td>0.303</td>
</tr>
<tr>
<td>Night-time SBP (mmHg)</td>
<td>139.16±3.19</td>
<td>140.18±3.16</td>
<td>134.66±2.66</td>
<td>0.392</td>
</tr>
<tr>
<td>Night-time DBP (mmHg)</td>
<td>90.56±2.91</td>
<td>90.82±3.59</td>
<td>84.50±2.85</td>
<td>0.256</td>
</tr>
<tr>
<td>Night-time SBP (mmHg)</td>
<td>11.91±0.80</td>
<td>12.43±1.90</td>
<td>17.47±1.61**</td>
<td>0.002</td>
</tr>
<tr>
<td>Night-time CV-SBP (mmHg)</td>
<td>8.55±0.53</td>
<td>9.02±0.80</td>
<td>12.92±1.13**</td>
<td>0.001</td>
</tr>
<tr>
<td>Night-time SD-SBP (mmHg)</td>
<td>9.65±0.33</td>
<td>10.13±0.70</td>
<td>11.48±1.16</td>
<td>0.258</td>
</tr>
<tr>
<td>Night-time CV-DBP (mmHg)</td>
<td>10.80±0.48</td>
<td>11.59±1.16</td>
<td>13.59±1.28</td>
<td>0.148</td>
</tr>
<tr>
<td>24-h SBP (mmHg)</td>
<td>124.96±3.94</td>
<td>128.17±4.36</td>
<td>122.89±2.99</td>
<td>0.618</td>
</tr>
<tr>
<td>24-h DBP (mmHg)</td>
<td>80.68±2.74</td>
<td>83.82±4.57</td>
<td>76.88±1.94</td>
<td>0.322</td>
</tr>
<tr>
<td>24-h SD-SBP (mmHg)</td>
<td>10.31±1.11</td>
<td>9.95±0.97</td>
<td>10.28±1.24</td>
<td>0.970</td>
</tr>
<tr>
<td>24-h CV-SBP (mmHg)</td>
<td>8.14±0.76</td>
<td>7.85±0.76</td>
<td>8.37±1.05</td>
<td>0.916</td>
</tr>
<tr>
<td>24-h SD-DPP (mmHg)</td>
<td>9.56±0.61</td>
<td>10.30±0.80</td>
<td>9.00±0.68</td>
<td>0.543</td>
</tr>
<tr>
<td>24-h CV-DPP (mmHg)</td>
<td>11.90±0.72</td>
<td>12.36±1.06</td>
<td>11.81±0.98</td>
<td>0.907</td>
</tr>
</tbody>
</table>

IL, interleukin; LVH, left ventricular hypertrophy; WBC, white blood cell; BMI, body mass index; TG, triglyceride; HDL-C, high-density lipoprotein-cholesterol; LDL-C, low-density lipoprotein-cholesterol; SBP, systolic blood pressure; DBP, diastolic blood pressure; SD, standard deviation; CV, coefficient of variation.

Post-hoc analysis: *p<0.05 compared with lower tertile; **p<0.01 compared with lower tertile.

Methods

Participants

Participants were recruited from the hypertension outpatient clinic at the Seoul National University Bundang Hospital. Candidates were those subjects who met the criteria of essential hypertension (systolic BP ≥140 mmHg or diastolic BP ≥90 mmHg).

Exclusion criteria were: subject is taking anti-hypertensive medication; subject has secondary hypertension, myocardiар infarction or had a cerebrovascular accident within the preceding 3 months; subject has clinically significant valvular heart disease, heart failure (class III, IV), renal insufficiency (serum creatinine ≥2.5 mg/dl), hepatic failure, uncontrolled diabetes mellitus and received drugs that could have affected the concentration of plasma cytokines (anti-inflammatory drugs, statins, and glitazones). Informed consent for the study was obtained from all patients.

Blood Sampling and ELISA

The blood sample from each fasted participant was obtained by venipuncture. Samples were placed into ethylene diamine tetra-acetic acid tubes. The tubes underwent centrifugation, and aliquots of plasma were then taken and stored at −70°C. Enzyme immunoassay for the quantitative determination of interleukin (IL)-6 and tumor necrosis factor (TNF)-α was performed using human cytokine enzyme immunoassay kits (Neogen Corporation, Lexington, KY, USA), according to the manufacturer’s instructions. The minimum detection limit of IL-6 and TNF-α were 3.4 pg/ml and 0.2 ng/ml, respectively. C-reactive protein (CRP) was measured using particle-enhanced light-scattering immunoassay (TBA-200FR system; Toshiba, Tokyo, Japan), according to the manufacturer’s instructions, and the lower detection limit was 0.01 mg/dl.

Ambulatory BP Measurement

Ambulatory BP was obtained using a non-invasive oscillometric system (P6 Pressurometer; Del Mar Reynold, CA, USA). The experienced technician placed an appropriately-sized BP cuff on the participant’s non-dominant arm and instructed the patient to go about his or her normal activities during the 24-h ambulatory BP monitoring period but refrain from vigorous physical activity. Before the start of the monitoring period, automatic readings were cross-checked against manually measured BP by auscultation to ascertain
that BP monitoring was correct. BP measurements taken 07:00h between 24:00h were regarded as ‘awake (daytime)’ measurements, and measurements taken between 24:00h and 07:00h were ‘asleep (night-time)’ measurements. The ambulatory monitor was programmed to record a subject’s BP every 30min during the awake period and every 60min during the asleep period. Recordings were excluded from the analysis when more than 15% of all readings were missing or incorrect. A participant’s BP variability was calculated as: 1) within-subject SD; and 2) the coefficient of variation (CV; CV = SD/mean value × 100%) of systolic and diastolic BP during each of the awake, asleep and 24-hour periods. Diurnal variation of BP was classified as being either non-dipper, dipper, extreme dipper and riser, as defined elsewhere.

### Statistical Analysis
Data were analyzed using SPSS ver.12 (SPSS Inc, Chicago, IL, USA). Continuous variables are expressed as the mean ± SEM and categorical variables are described in terms of frequencies and percentages. Comparisons among each of the IL-6, TNF-α, and high sensitive-CRP (hs-CRP) tertiles were performed using one-way analysis of variance, followed by application of the Bonferroni method for multiple comparisons. Any association between inflammatory marker and ambulatory BP index was tested by calculating bivariate Pearson’s correlation coefficients. Stepwise multiple linear regression models were applied to examine whether the inflammatory markers were related to BP or BP variability, after adjustment for age, gender, body mass index, smoking, and target organ damage (left ventricular hypertrophy (LVH), albuminuria). A p-value less than 0.05 denoted the presence of a statistically significant difference.

### Results
A total of 55 hypertensive patients who met the criteria were recruited. Of these 55 subjects, 52 patients had complete data for all variables of interest and formed the study population. The average age was 55.9±1.5 years (range, 30–78 years) and 54% were male. The mean systolic and diastolic office BP of the participants was 160.0±2.1 mmHg and 95.2±1.5 mmHg, respectively.

Participants were grouped into tertiles, according to their plasma IL-6, TNF-α, and hs-CRP level. A significant positive association between age and IL-6 level was identified (Table 1). The mean age in the lower tertile of IL-6 was 56.1±2.55 and upper tertile (60.1±3.25) for (P for trend = 0.038).

### Table 2 Clinical, Laboratory, and Hemodynamic Characteristics According to the TNF-α Tertile

<table>
<thead>
<tr>
<th>TNF-α tertile</th>
<th>P for trend</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤11.34</td>
<td></td>
</tr>
<tr>
<td>11.34–19.95</td>
<td></td>
</tr>
<tr>
<td>&gt;19.95</td>
<td></td>
</tr>
</tbody>
</table>

Age (years)     | 61.76±2.34  | 53.88±2.24  | 52.50±2.69** | 0.020 |
Male gender     | 41.2%       | 55.6%       | 64.7%        | 0.382 |
Smoking         | 5.9%        | 11.8%       | 25.0%        | 0.271 |
LVH             | 11.8%       | 11.8%       | 31.3%        | 0.247 |
Albuminuria     | 0%          | 17.6%       | 25.0%        | 0.102 |
WBC (10³/μl)    | 5.76±0.34   | 6.31±0.48   | 5.56±0.33    | 0.380 |
Hemoglobin (g/dl)| 13.94±0.36  | 14.50±0.46  | 14.93±0.27   | 0.167 |
Platelet (10³/μl)| 233.41±12.08| 218.56±10.23| 241.00±6.70  | 0.782 |
BMI (kg/m²)     | 24.27±0.52  | 26.63±0.67* | 24.88±0.52   | 0.027 |
Glucose (mg/dl) | 101.25±2.28 | 105.50±3.29 | 96.13±2.79   | 0.073 |
Total cholesterol (mg/dl) | 225.53±7.45 | 210.44±5.27 | 207.44±9.80  | 0.212 |
TG (mg/dl)      | 113.20±8.07 | 132.31±16.04| 117.63±13.86 | 0.571 |
HDL-C (mg/dl)   | 67.53±5.97  | 58.00±2.87  | 67.19±4.10   | 0.233 |
LDL-C (mg/dl)   | 136.56±8.39 | 125.97±5.46 | 116.73±8.93  | 0.208 |
Office SBP (mmHg)| 159.41±2.96 | 159.76±5.53 | 162.50±3.84  | 0.828 |
Office DBP (mmHg)| 89.65±2.26  | 96.12±2.12  | 100.63±2.69* | 0.008 |

day-time
SBP (mmHg)   | 132.44±2.62 | 141.06±2.61 | 145.09±2.97**| 0.007 |
DBP (mmHg)   | 81.79±2.51  | 92.71±2.31* | 94.48±3.09** | 0.003 |
SD-SBP (mmHg) | 15.56±1.84  | 13.77±0.99  | 13.63±1.06   | 0.542 |
CV-SBP (mmHg) | 11.73±1.35  | 9.81±0.72   | 9.42±0.74    | 0.214 |
SD-DBP (mmHg) | 10.31±1.22  | 10.75±0.56  | 10.37±0.48   | 0.921 |
CV-DBP (mmHg) | 12.64±1.39  | 11.75±0.73  | 11.29±0.84   | 0.647 |

Night-time
SBP (mmHg)   | 119.60±3.15 | 131.26±3.42 | 128.95±3.62  | 0.043 |
DBP (mmHg)   | 73.17±2.17  | 86.85±2.27* | 83.53±3.48*  | 0.002 |
SD-SBP (mmHg) | 11.08±1.63  | 11.59±1.06  | 9.27±1.27    | 0.359 |
CV-SBP (mmHg) | 9.17±1.23   | 9.12±0.82   | 7.13±0.91    | 0.279 |
SD-DBP (mmHg) | 8.54±0.76   | 11.15±0.61* | 9.21±0.67    | 0.028 |
CV-DBP (mmHg) | 11.84±1.18  | 12.89±0.71  | 11.06±0.75   | 0.375 |
24-h
SBP (mmHg)   | 130.49±2.47 | 139.11±2.56 | 141.69±2.87* | 0.011 |
DBP (mmHg)   | 80.47±2.37  | 91.88±2.09**| 92.16±3.05** | 0.002 |
SD-SBP (mmHg) | 14.93±1.11  | 14.47±1.06  | 15.12±1.11   | 0.910 |
CV-SBP (mmHg) | 11.45±0.78  | 10.47±0.80  | 10.69±0.73   | 0.646 |
SD-DBP (mmHg) | 10.24±0.88  | 11.40±0.48  | 11.57±0.50   | 0.158 |
CV-DBP (mmHg) | 12.76±0.64  | 12.52±0.60  | 12.77±0.71   | 0.953 |

TNF, tumor necrosis factor. Other abbreviations as given in Table 1.
Post-hoc analysis: *p<0.05 compared with lower tertile; **p<0.01 compared with lower tertile.
Quite the opposite, whereby the mean age was lowest in the upper tertile (12.92±1.3 mmHg) was even higher than the lower (8.55±0.53 mmHg) and middle tertile (9.02±0.79 mmHg) during the awake period (P for trend=0.001).

Meanwhile, the association between TNF-α and age was quite the opposite, whereby the mean age was lowest in the upper tertile group (Table 2). Systolic and diastolic BPs were higher in the middle and upper tertile groups compared with the lower tertile group. By contrast, there was no difference in BP and BP variability index among the hs-CRP tertile groups.

The association between inflammation and pattern of diurnal variation was investigated (Table 3). White blood cell count and hs-CRP concentration were significantly increased in the riser group. However, IL-6 and TNF-α levels did not differ among the different diurnal variation groups.

Correlation analysis showed different associations for IL-6 and TNF-α. TNF-α level correlated significantly with BP index, whereas IL-6 level correlated with BP variability index. There was no significant correlation between hs-CRP and either BP or BP variability index (Table 4).

The interaction between inflammatory marker and either BP or BP variability was still significant even after adjusting for the effect of age, gender, body mass index, smoking, and target organ damage in a stepwise multiple linear regression model (Table 5). The SD of daytime systolic BP (I=0.065, p=0.001) and age (I=0.024, p=0.016) were all positively and significantly related to IL-6. In contrast, only daytime diastolic BP (I=0.029, p=0.002) was independently related to TNF-α.

Discussion

In the present study, we investigated the link between BP variability and inflammatory marker in hypertensive patients. We showed that there is a significant association between BP variability index and inflammatory marker, especially IL-6 level, among the study’s participants. Because of the cross-sectional study design, we cannot tell the causality of these 2 parameters. However, the link between BP variability index and inflammatory marker may suggest the interactive effects of inflammation and BP variation in the process of target organ damage.

Previous studies have shown that 24-h BP variability has clinical relevance in hypertensive patients. There are posi-
active independent associations between target organ damage (such as LVH or carotid atherosclerosis) and BP variability. Furthermore, it has been reported that cardiovascular events are greater in patients with a wider BP variability than in those with a narrower BP variability.

However, the exact mechanisms explaining the clinical significance of BP variability are, as yet, unclear. One of the putative mechanisms is hemodynamic stress on the vessel wall. Steeper BP variations may produce a greater stress on the vessel wall and, consequently, result in medial hypertrophy of the large arteries. This finding indicates that the alteration of vessel wall tension associated with steeper BP increases and reductions may initiate medial hypertrophy and early atherosclerosis formation in the arterial wall of large vessels. Thus, target organ damage in essential hypertension, in addition to the level of BP values and the magnitude of BP fluctuations, also may be related to BP changes occurring with a greater rate.

Inflammation theory is another promising hypothesis. A previous experimental study with sinoaortic denervation (SAD) rats, which is the animal model for BP variation, showed that high BP variability is related to organ damage. Inflammation-related factors are increased in SAD rats, with anti-inflammatory and antioxidant treatment reducing the organ damage induced by SAD. Hence, it is proposed that inflammation is one of the mechanisms underlying organ damage in SAD rats. In addition, impaired endothelial function and increased neointimal formation after balloon injury were observed in the SAD model, implying that increased BP variability, independent of the average BP level, may contribute to the progression of atherosclerosis.

In the present study, we observed an association between BP variability index and inflammatory markers, such as TNF-α and IL-6, both of which are the key molecule of inflammatory process. Some reports have shown a significant association between inflammatory marker and elevated BP in apparently healthy patients. In addition, CRP level is associated with future development of hypertension, which means that hypertension is, in part, an inflammatory disorder. Moreover, CRP and BP are both independent determinants of future cardiovascular disease, and their predictive value is additive.

However, there was little evidence to demonstrate an association between inflammation and BP variability in human subjects. Recently, Abramson et al demonstrated positive associations between markers of inflammation and BP variability in healthy, normotensive adults. Nonetheless, the interaction between BP variability and inflammation in hypertensive patients has never been investigated.

In the present study, each inflammatory marker showed a different association with either BP or BP variability index. There was no association between hs-CRP and either BP or BP variability index. In contrast, there was a significant association between IL-6 and BP variability, and between TNF-α and BP variability index. The reason(s) for the different associations observed requires further investigation.

It is well known that IL-6 and TNF-α both induce the expression of CRP in liver. Accordingly, increased levels of IL-6 and TNF-α are required for the expression of CRP. In other words, CRP is a less sensitive inflammatory marker than either IL-6 or TNF-α. In the present study, the CRP concentration was 0.01 mg/dl (lower detection limit) in 23 patients (44.2%). As a result, the hs-CRP tertile groups were not distributed equally in our study. Consequently, the lack of association between CRP and either BP or BP variability index in this study may be because the sample size was too small. Furthermore, various inflammation processes might be involved in the increased expression of CRP. Accordingly, the different associations between specific inflammatory markers and either BP or BP variability index could be explained by such characteristics.

The present study has several limitations. Because of the cross-sectional study design, we cannot tell the causality of BP variability and inflammation. However, the link between BP variability index and inflammatory marker suggests that the underlying mechanism may be a result from the process of target organ damage. To identify the causal relationship between BP variability and inflammation, a long-term follow-up study or well-designed intervention study is needed. Our study was based on a relatively small sample, so it is unclear whether the results can be applied to other populations. Although the autonomic nervous system, the renin–angiotensin system, and salt sensitivity also have a great impact on BP variability, we could not evaluate the effect of any other system that might be involved in BP variability.

Despite these limitations, we believe that the study provides new scientific information because, to our knowledge,
it is the first study to report statistically significant positive associations between inflammatory markers and BP variability in hypertensive patients.

References


