

Elevated Endothelin-1 Levels Are Associated With Decreased Arterial Elasticity in Hypertensive Patients

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Endothelin-1 stimulates collagen synthesis and is increased in hypertension, but its effect on collagen degradation remains unknown. The current study tested the hypothesis that elevated endothelin-1 levels are associated with decreased collagenase activity, markers of collagen degradation, and arterial compliance in hypertensive patients. Normotensive (n=10) and hypertensive (n=13) patients who were not on any antihypertensive medication were recruited, and small and large artery elasticity index, systemic vascular resistance, pulse pressure, and blood pressure were determined using blood pressure waveform analysis. Large artery elasticity index and collagen degradation products were decreased whereas endothelin-1, systemic vascular resistance, and pulse pressure were elevated in hypertensive patients. Plasma endothelin-1 was negatively correlated with a cross-linked C-terminal telopeptide of collagen type I, a collagen degradation marker (r=-0.43; p=0.04), collagenase matrix metalloproteinase-1 (r=-0.48; p=0.02), and large artery elasticity (r=-0.45; p=0.03) and positively

correlated with pulse pressure (r=0.68; p=0.0005). These results suggest that endothelin-1 contributes to decreased arterial compliance in hypertension via inhibition of collagen degradation. (J Clin Hypertens. 2006;8:549-554) ©2006 Le Jacq

Arterial hypertension is associated with increased risk of target organ damage, resulting in stroke, left ventricular hypertrophy, and coronary artery disease.¹ A number of observational and clinical studies, including the Framingham Heart Study, demonstrated that there is a strong association among blood pressure (BP), pulse pressure (PP), and adverse cardiac events.²⁻⁵ Since BP and PP are related to the physical properties of elastic arteries, arterial stiffness and pulse wave velocity are now considered cardiovascular risk factors; assessment of arterial stiffness may improve efforts for optimal treatment of high BP and related complications.^{5,6} In recent years, the development of noninvasive techniques such as brachial artery pulse wave analysis has enabled the rapid assessment of arterial stiffness.⁷⁻⁹

Structural changes in the vessel wall such as increased extracellular matrix deposition (especially collagen type I), resulting in arterial fibrosis, contribute to increased stiffness.¹⁰⁻¹² Enhanced collagen deposition may be due to increased collagen synthesis and/or decreased degradation by matrix metalloproteinases (MMP), including collagenase MMP-1. Laviades and colleagues^{13,14} reported that plasma levels of MMP-1 and a cross-linked C-terminal telopeptide of collagen type I (ICTP), a collagen type I degradation product, were decreased in patients with essential hypertension

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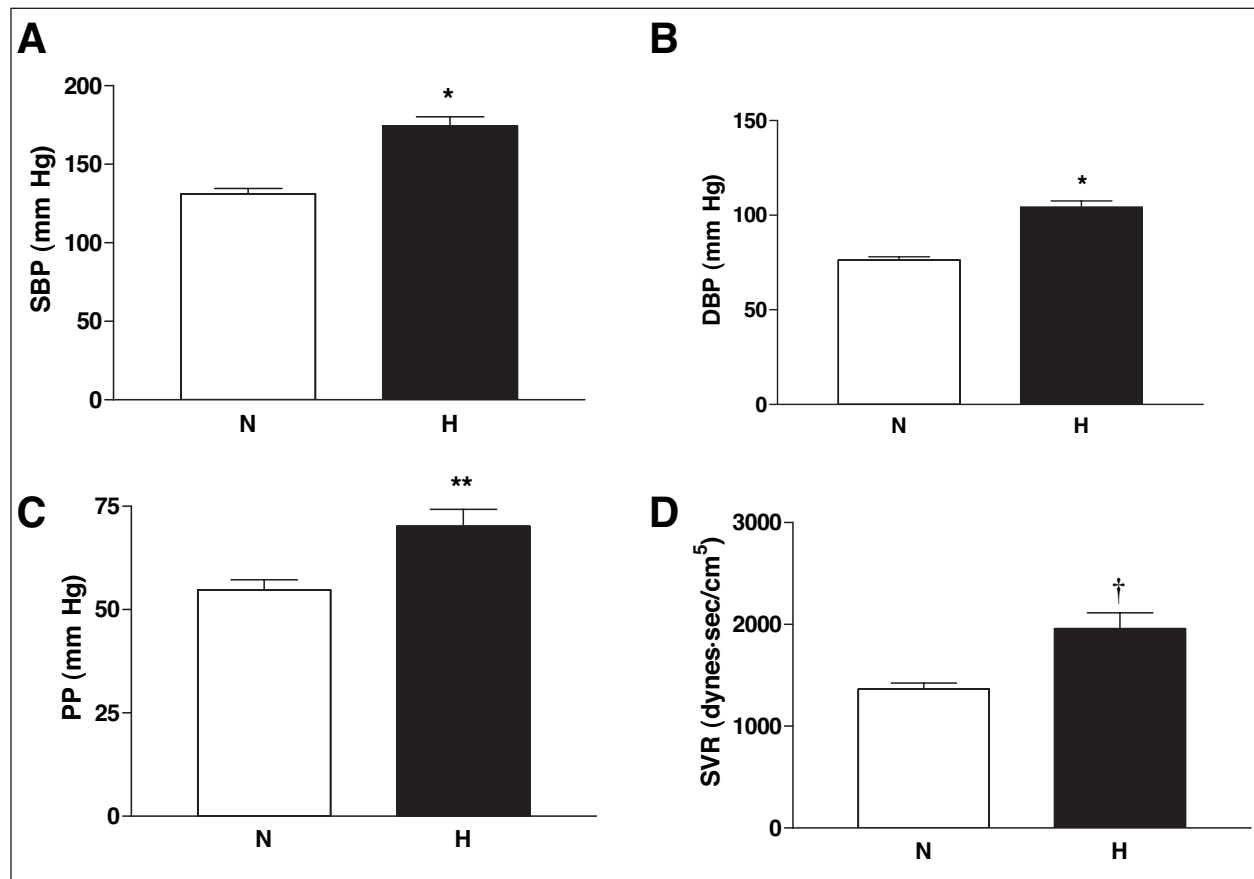


Figure 1. Comparison of hemodynamic parameters in normotensive (N) and hypertensive (H) patients. Systolic blood pressure (SBP) (A), diastolic blood pressure (DBP) (B), pulse pressure (PP) (C), and systemic vascular resistance (SVR) (D) were significantly elevated in the hypertensive group. Results are given as mean \pm SEM. * $p < 0.001$; ** $p = 0.008$; † $p = 0.003$ vs. normotensive

Table. Patient Demographics	NORMOTENSIVE	HYPERTENSIVE
	(N=10)	(N=13)
Age (yr) (mean \pm SD)	36 \pm 6	40 \pm 3
Race (African American/ Caucasian)	3/7	8/5
Sex (female/male)	6/4	8/5
Body mass index (kg/m ²) (mean \pm SD)	27 \pm 2	30 \pm 2

and left ventricular hypertrophy, whereas levels of endogenous inhibitors of matrix proteinases were increased. Using human arterial tissue, our group demonstrated that not only MMP-1 and MMP-9, but MMP inducer and activator proteins, are also down-regulated in the hypertensive state.¹⁵ However, the relationship of circulating levels of collagen synthesis and degradation products such as ICTP, as well as collagenase levels to arterial stiffness remained unknown.

While increased stiffness is mainly attributed to structural changes in the vessel wall, changes in smooth muscle tone also affect the stiffness of

arteries. Vasoactive peptides such as endothelin-1 (ET-1) and angiotensin II are involved in this process.⁸ McEniery et al.¹⁶ reported that endogenous ET-1 regulates arterial pulse wave velocity. In addition to vasoconstrictor effects, ET-1 stimulates collagen synthesis. We and others have shown that ET receptor antagonism improves extracellular matrix deposition in experimental hypertension.^{11,12,17-21} Thus, ET-1 is an important vasoactive factor that can affect both functional and structural arterial stiffening. Accordingly, the goal of this study was to study the relationship among circulating ET-1, markers of vascular remodeling, and indices of arterial elasticity in normotensive vs. hypertensive patients.

MATERIALS AND METHODS

Patient Enrollment and Pulse Contour Analyses

The study protocol was approved by the Human Assurance Committee at the Medical College of Georgia. Ten normotensive and 13 hypertensive patients who were not receiving any antihypertensive medication were recruited from the hypertension clinic. BP >160 mm Hg systolic and/or

95 mm Hg diastolic was considered hypertensive. Exclusion criteria included renal failure (serum creatine >2 mg/dL), pulmonary edema, asthma (exacerbation within the past month), recent myocardial infarction (<6 months) or angina (<1 month), cardiogenic shock, and heart failure. Demographics of patients are summarized in the Table. After patient consent was obtained, pulse contour analysis was measured using a CVProfilor DO-2020 (Hypertension Diagnostics, Inc., Eagan, MN) cardiovascular profiling system. Patients remained supine for 5 minutes before measurement of arterial elasticity. The BP cuff was placed on the left upper arm. A wrist stabilizer was positioned on the patient's right wrist to stabilize the position of the radial artery. The arterial sensor was placed over the right radial artery at the point of maximal pulsation to capture an analog BP waveform data signal and adjusted to the highest relative signal strength. Three measurements approximately 1 minute apart were performed.

Blood Chemistry

Upon completion of compliance measurements, blood was collected in ethylenediaminetetraacetic acid tubes. Plasma ET-1, MMP-1, and collagen degradation and synthesis products ICTP and procollagen type I N-terminal peptide, respectively (DiaSorin, Stillwater, MN), were determined as we have described previously.¹⁵

Data Analyses

Comparisons between hypertensive and nonhypertensive groups were made using Student *t* tests. Pearson correlations were used to assess the linear association between plasma ET-1 levels and other variables. Statistical significance was set at $p < 0.05$. SAS version 8.2 (SAS Institute, Inc., Cary, NC) was used for all analyses.

RESULTS

There were significant differences only in systolic and diastolic BP, PP, and systemic vascular resistance between normotensive and hypertensive patients, which showed a significant increase in hypertensive patients as expected (Figure 1). Large artery elasticity index was significantly decreased ($p = 0.043$), and there was a trend for decreased small artery elasticity index in hypertensive patients ($p = 0.11$) (Figure 2).

Circulating markers of vascular remodeling were also analyzed. Collagen degradation marker ICTP was significantly decreased in hypertensive patients ($p = 0.005$), but there was no change in a collagen

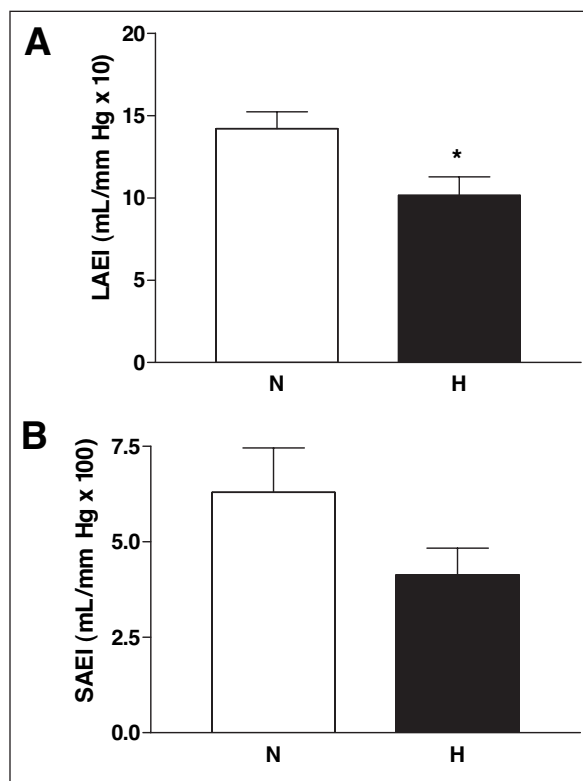


Figure 2. Arterial elasticity measurements in normotensive (N) and hypertensive (H) patients. Large artery elasticity index (LAEI) (A) is decreased and there is a trend for decreased small artery elasticity index (SAEI) (B) in the hypertensive group. Results are given as mean \pm SEM. * $p = 0.043$ vs. normotensive

synthesis marker between groups (Figure 3A and B). Consistent with decreased collagen markers, fibrillar collagenase MMP-1 (Figure 3C) was also decreased in hypertensive patients ($p < 0.001$). Collagen degradation levels showed an inverse relationship with increased systolic BP ($r = -0.57$; $p = 0.006$), diastolic BP ($r = -0.55$; $p = 0.008$), and PP ($r = -0.48$; $p = 0.024$). Decreased matrix levels were also associated with increased systolic BP ($r = -0.67$; $p = 0.0006$), diastolic BP ($r = -0.72$; $p = 0.0002$), and PP ($r = -0.49$; $p = 0.021$).

As shown in Figure 3D, plasma ET-1 levels were also elevated in the hypertensive group ($p = 0.018$). Pearson correlations demonstrated that plasma ET-1 was negatively correlated with collagen degradation product ICTP ($r = -0.43$; $p = 0.04$), collagenase MMP-1 ($r = -0.48$; $p = 0.02$), and large vessel elasticity ($r = -0.45$; $p = 0.03$), and positively correlated with PP ($r = 0.68$; $p = 0.0005$).

DISCUSSION

The important findings of this study are that collagen degradation, and not synthesis, is altered in association with elevated ET-1 levels, and that

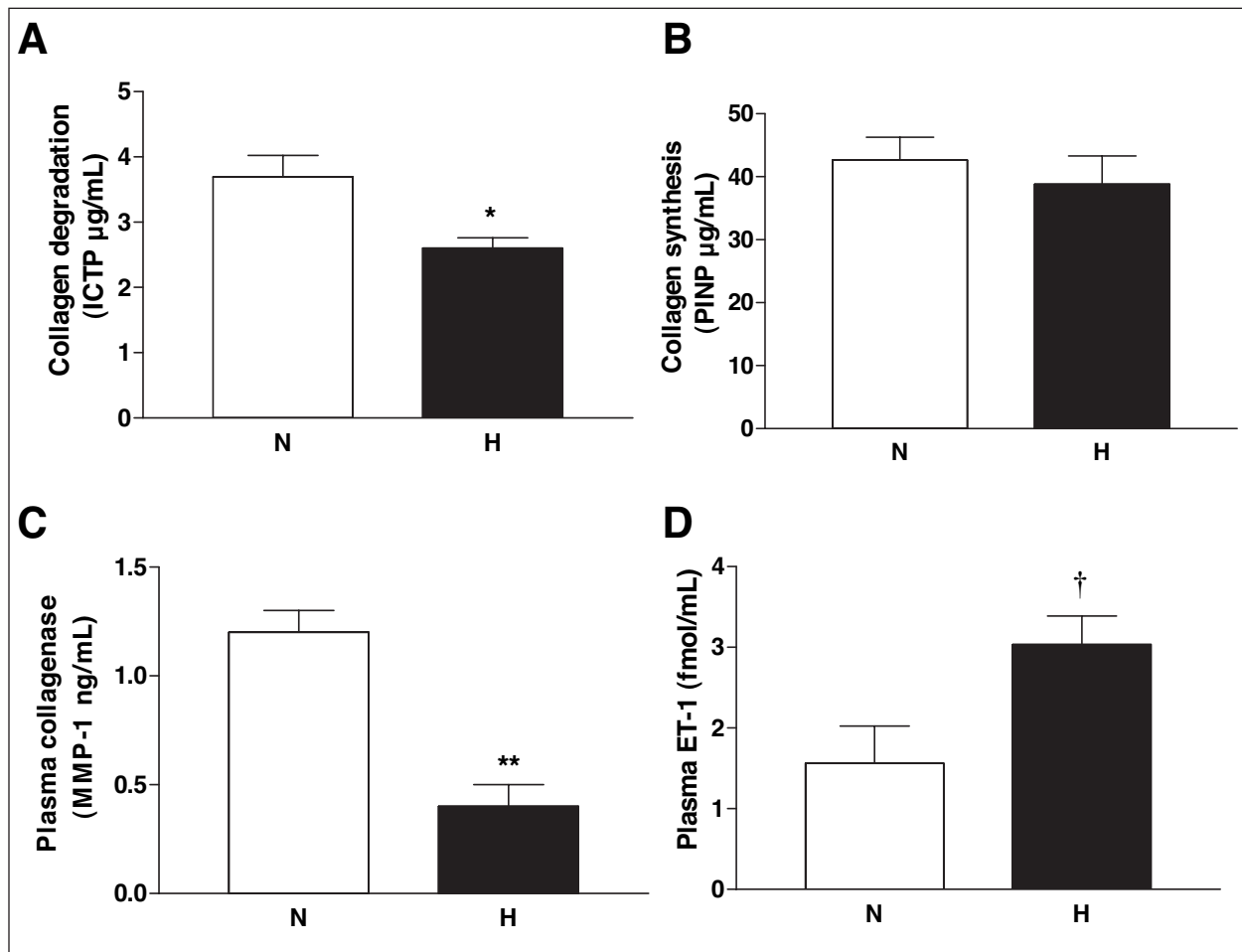


Figure 3. Plasma levels of vascular remodeling markers and endothelin-1 (ET-1) in normotensive (N) and hypertensive (H) patients. Collagen degradation product cross-linked C-terminal telopeptide of collagen type I (ICTP) and collagenase matrix metalloproteinase (MMP-1) are decreased, whereas ET-1 levels are elevated in hypertensive patients. Results are given as mean \pm SEM. * $p=0.005$; ** $p<0.001$; † $p=0.018$ vs. normotensive

elevated ET-1 levels are associated with decreased large vessel elasticity and increased PP in hypertensive patients. These results suggest that ET-1 contributes to decreased arterial elasticity in hypertension via inhibition of collagen degradation.

It has long been recognized that PP is an indicator of arterial stiffness and that arterial stiffness is an important predictor of cardiovascular risk.²⁻⁵ Increased arterial stiffness results in decreased vascular compliance, which reflects the inability of vessels to adapt to changes in BP. Therefore, in recent years, evaluation of arterial compliance in addition to BP measurements has been recommended by some investigators not only to monitor the effectiveness of antihypertensive therapy but also to determine early vasculopathy.^{7,8,22-25} We and others have shown that both large and small artery elasticity can be easily assessed by noninvasive pulse wave analysis.^{7,9,22} The current study provided evidence that large artery elasticity is significantly decreased in patients

whose BP is not currently under control. Decreased arterial elasticity may arise from pathologic vascular remodeling of arteries in hypertension, which involves growth, extracellular expansion, and fibrosis.²⁶⁻²⁸ Alterations in the abundance and properties of structural elements such as collagen and elastin are important determinants of vascular stiffness. MMPs regulate this process in a complex manner by degrading matrix proteins such as collagen and elastin.²⁹ In our previous study, we demonstrated that both inducer and activator proteins of the arterial MMP system are reduced in hypertension, leading to enhanced matrix deposition.¹⁵ The current study provides evidence that circulating MMP-1 levels are also decreased in hypertensive patients and that this is correlated with decreased collagen degradation markers as well as decreased arterial elasticity.

Several other studies have reported changes in the soluble markers of vascular remodeling in hypertension.^{13,14,30-32} A recent report provided

evidence that 6-month treatment with felodipine restored decreased MMP levels in hypertensive patients, whereas diltiazem had no effect.³² Yasmin et al.³³ investigated plasma proteinase levels in isolated systolic hypertension as well as in healthy individuals and found that aortic stiffness is related to these not only in patients with systolic hypertension but also in healthy individuals—indicating the importance of this system in structural properties of the vessel wall. The current study complements these findings and provides evidence that decreased circulating MMP-1 levels in hypertensive patients are associated with increased arterial stiffness.

In addition to the structural properties of the vasculature, it is reasonable to speculate that smooth muscle tone can also affect arterial stiffness. Early studies suggested that ET-1, the most potent vasoconstrictor with mitogenic and profibrotic properties, contributes to basal tone of large arteries, and that circulating ET-1 levels are correlated with decreased arterial elasticity.^{34,35} ET-1 is also known to stimulate the synthesis of collagen. In an experimental model of borderline hypertension, we reported that ET-1 regulates the activation of MMPs in the aorta.¹⁷ These past studies suggest that ET-1 regulates MMP expression/activity, leading to augmented collagen deposition in a complex and time-dependent manner.³⁶ The current study extends the findings obtained with experimental models to clinical studies and demonstrates that elevated ET-1 levels are associated with decreased arterial elasticity, plasma MMP-1, and collagen degradation markers in hypertensive patients. As we previously reported, plasma ET-1 levels are elevated to a greater degree in African-American patients; in this study there are more African-American individuals in the hypertensive group than in the normotensive group.³⁷ Whether this imbalance contributes to the findings of increased artery stiffness in the hypertensive group remains to be determined.

Perspectives

There are several limitations of our study that should be recognized. First, the results of this study do not provide evidence of a cause-and-effect relationship between increased arterial stiffness and ET-1 or MMPs. The current findings suggest that elevated ET-1 levels down-regulate collagenase expression/activity, which results in decreased collagen turnover and, ultimately, increased arterial stiffness. Second, we evaluated only collagenase- and gelatinase-type MMPs; other factors in this group remain to be studied. Third, we measured

the indices of vascular compliance and plasma markers at only one time point. Hypertensive patients who participated in the study were not receiving any antihypertensive treatment when they were enrolled in the study. However, there is limited information regarding the duration of hypertension or the previous use of antihypertensive medications in this cohort. Finally, the number of subjects is small. Nevertheless, the importance of the current results is three-fold. First, recent studies have identified plasma MMP-9 as a novel predictor of cardiovascular mortality.^{38–40} Thus, evaluation of MMPs involved in vascular remodeling may serve as a marker of vascular compliance and cardiovascular risk. Second, assessment of arterial stiffness may be important for the management of hypertension and may provide a noninvasive means to monitor BP control. The third and the most speculative interpretation of the current data is that ET receptor antagonism may provide BP control and vascular protection by improving arterial elasticity in hypertensive patients.

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