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Citation for the original published paper (version of record):

Carlsson, A., Ruge, T., Sundström, J., Ingelsson, E., Larsson, A. et al. (2013)

Association between circulating endostatin, hypertension duration, and hypertensive target-organ damage.

*Hypertension*, 62(6): 1146-1151

<http://dx.doi.org/10.1161/HYPERTENSIONAHA.113.02250>

Access to the published version may require subscription.

N.B. When citing this work, cite the original published paper.

Permanent link to this version:

<http://urn.kb.se/resolve?urn=urn:nbn:se:uu:diva-211651>

# **The association between circulating endostatin, hypertension duration and hypertensive target organ damage**

Axel C Carlsson<sup>1,2\*</sup>, Toralph Ruge<sup>2, 3\*</sup>, Johan Sundström<sup>4</sup>, Erik Ingelsson<sup>5</sup>, Anders Larsson<sup>4</sup>, Lars Lind<sup>4</sup>, and Johan Ärnlöv<sup>2,6\*\*</sup>

## **Running short title: Endostatin, hypertension duration and organ damage**

<sup>1</sup>Centre for Family Medicine, Department of Neurobiology, Care Sciences and Society, Karolinska Institutet, Huddinge, Sweden

<sup>2</sup>Department of Public Health and Caring Sciences/ Section of Geriatrics Uppsala University, Uppsala Sweden

<sup>3</sup>Department of Surgery, Umeå University, Umeå, Sweden

<sup>4</sup>Department of Medical Sciences, Uppsala University Hospital, Uppsala, Sweden

<sup>5</sup>Department of Medical Epidemiology and Biostatistics, Karolinska Institutet, Stockholm, Sweden

<sup>6</sup>School of Health and Social Studies, Dalarna University, Falun, Sweden

\*Contributed equally to this manuscript (alphabetical order)

Corresponding author: Axel C. Carlsson

E-mail: axelcefam@hotmail.com

And

Johan Ärnlöv, Department of Public Health and Caring Sciences/Geriatrics, Uppsala Science Park, SE-75185 Uppsala, Sweden, Fax: +46-18-6117976; phone: +46-70-6077703

E-mail: johan.arnlov@pubcare.uu.se

Words in abstract: 250

Number of words in main text: 2803

## **Abstract**

To study associations between circulating endostatin, hypertension duration, and hypertensive target organ damage.

Long-lasting hypertension induces cardiovascular and renal remodelling. Circulating Endostatin, a biologically active derivate of collagen XVIII, has been suggested to be a relevant marker for extracellular matrix turnover and remodelling in various disease. Yet, the role of endostatin in hypertension and hypertensive target organ damage is unclear.

Serum Endostatin was measured in 2 independent community-based cohorts; the Prospective Investigation of the Vasculature in Uppsala Seniors (PIVUS; women 50%, n=812, mean age 75 years) and the Uppsala Longitudinal Study of Adult Men (ULSAM, n=785, mean age 77 years). Retrospective data on blood pressure measurements and antihypertensive medication (PIVUS >5 years, ULSAM >27 years), and cross-sectional data on echocardiographic left ventricular mass, endothelial function (endothelial dependent vasodilation assessed by the invasive forearm model), and urinary albumin/creatinine-ratio was available. In PIVUS, participants with  $\geq 5$ -years history of hypertension portrayed 0.42 SD (95% CI 0.23-0.61,  $p < 0.001$ ) higher serum endostatin, compared to normotensives. This association was replicated in ULSAM, where participants with 27 years hypertension duration had highest endostatin (0.57 SD higher, 95% CI 0.35-0.80,  $p < 0.001$ ). In addition, higher endostatin was associated with higher left ventricular mass, worsened endothelial function, and higher urinary albumin/creatinine-ratio ( $p < 0.03$  for all) in participants with prevalent hypertension.

Circulating endostatin is associated with the duration of hypertension, and vascular, myocardial and renal indices of hypertensive target organ damage. Further studies are warranted to assess the prognostic role of endostatin in hypertensive individuals.

**Key words:** arterial stiffness, endothelial dysfunction, vascular remodelling, remodelling of extracellular matrix, angiogenesis, anti-angiogenesis, population based studies, epidemiology

## **Introduction**

Hypertension is a major cause of cardiovascular disease and was recently put forward as the leading risk factor for global disease burden.<sup>1</sup> Long standing hypertension induces damage to the vasculature and myocardium,<sup>2</sup> as well as to the kidneys.<sup>3</sup> An increased turnover of the extra cellular matrix has been suggested as a key element in the underlying pathophysiology of this hypertensive target organ damage.<sup>4</sup> Yet to date, relevant endogenous biomarkers mirroring hypertension related cardiovascular remodelling are scarce.

Endostatin is produced by the proteolytic cleavage of the c-terminal domain of collagen XVIII, a component of the extra cellular matrix.<sup>5</sup> Release of endostatin is mainly induced by the stimulation of elastase, metalloproteinases (MMPs) and cathepsins.<sup>6-8</sup> Endostatin plays a role in the local balance of angiogenesis as a potent inhibitor and has been suggested to be of particular importance in the growth and spreading of malignant diseases.<sup>9-12</sup> The role of endostatin in the development of cardiovascular diseases, however, is less studied.

In short, Collagen XVIII increases in myocardial and renal tissues as well as the arterial wall with hypertension.<sup>4, 13, 14</sup> Meanwhile, extracellular matrix proteinases also increase in those tissues<sup>4, 17, 18</sup> Thus, endostatin, a cleaved product of collagen XVIII, could be an indicator of adverse extracellular remodelling in hypertension.

In the present study, we hypothesized that long-term exposure to hypertension induces extracellular matrix remodelling of the cardiovascular tree which is reflected by elevated levels of circulating endostatin. Accordingly, we aimed to study the association between duration of hypertension and circulating endostatin levels, in 2 community-based cohorts of elderly. As a second step, we also wanted to investigate the cross-sectional association between endostatin and vascular, myocardial and renal indices of hypertensive target organ

damage, namely endothelial dependent vasodilation, left ventricular mass, and urinary albumin/creatinine ratio.

## Methods

### Study populations

#### *The Prospective Investigation of the Vasculature in Uppsala Seniors (PIVUS)*

All 70-year old men and women, living in Uppsala Sweden, between 2001-2004, were eligible for the PIVUS study<sup>15</sup> (described in detail on <http://www.medsci.uu.se/pivus/pivus.htm>). Of 2025 invited individuals, 1016 agreed to participate. A second examination cycle was performed after 5 years when participants were approximately 75 years old (2006-2009). Of 964 invited individuals, 827 agreed to participate at this follow-up. Of these, 15 participants were excluded due to missing data on endostatin leaving 812 participants as the study sample. This second examination cycle was used as the baseline for the retrospective analyses between endostatin and the duration of hypertension and for the cross-sectional association between endostatin and urinary albumin/creatinine ratio while the cross-sectional association between endostatin and left ventricular mass and endothelial function was assessed at the first examination cycle as these data was not available at the second examination cycle.

#### *The Uppsala Longitudinal Study of Adult Men (ULSAM)*

The ULSAM study was initiated in 1970. All 50-year-old men, born in 1920-24 and living in Uppsala, Sweden, were invited to a health survey, focusing at identifying cardiovascular risk factors<sup>16</sup> (described in detail on <http://www.pubcare.uu.se/ULSAM>). These present analyses are based on the fourth examination cycle, when participants were approximately 77 years old (1998-2001). Of 1398 invited men, 838 participated. Of these, 53 were excluded due to missing data on endostatin leaving 785 participants as the present study sample.

All participants in both studies gave written informed consent and the Ethics Committee of Uppsala University approved the study protocols.

## **Baseline investigations**

The investigations in PIVUS and ULSAM were performed using similar standardized methods, including anthropometrical measurements, blood pressure, blood sampling, and questionnaires regarding socioeconomic status, medical history, smoking habits, medication and physical activity level.<sup>15, 16</sup> Venous blood samples were drawn in the morning after an overnight fast and stored at  $-70^{\circ}\text{C}$  until analysis. Serum levels of endostatin were analyzed using a commercially available ELISA kit for endostatin (DY1098, R&D Systems, Minneapolis, MN). The assays had a total coefficient of variation (CV) of approximately 7%.

Diabetes mellitus was diagnosed as fasting plasma glucose  $\geq 7.0$  mmol/l ( $\geq 126$ mg/dl), or use of anti-diabetic medication. Prevalent cardiovascular disease at baseline was defined as a history of ischemic heart disease or cerebrovascular disease, or Q-, QS-complexes or left bundle-branch block in baseline ECG. Leisure time physical activity was assessed by a questionnaire as previously described.<sup>17</sup>

Hypertension was defined as having a systolic/diastolic blood pressure above or equal to 140/90 mmHg and/or antihypertensive treatment. In PIVUS, we used two different ways to assess the hypertension duration. First, the duration of hypertension was assessed at the second examination cycle, based on blood pressure measurements and the use of antihypertensive medication at the first examination cycle 5 years earlier. Second, data on self-reported duration of hypertension, based on a questionnaire, was also used. The participants with hypertension were asked for how many years they had been diagnosed with hypertension. In the ULSAM cohort, the first approach was used with retrospective data on hypertension status, based on blood pressure measurements and use of antihypertensive medication, at examinations 6, 17, and 27 years prior to the baseline of examination cycle 4.

At the first examination cycle of PIVUS, forearm blood flow (FBF) was measured by venous occlusion plethysmography (Elektromedicin, Kullavik, Sweden) with the strain-gauge technique. An arterial cannula was placed in the brachial artery. Resting FBF was measured 30 min after cannula insertion, and local intra-arterial drug-infusions were given during 5 minutes for each dose with a 20 minutes washout period between the drugs. The infused dosages were 25 and 50 ug/minute for Acetylcholine (Clin-Alpha, Switzerland) and 5 and 10 ug/minute for SNP (Nitropress, Abbot, UK). The drugs were given in a random order at a maximal rate of 1 ml/min. Endothelium-dependent vasodilation with this technique (EDV) was defined as FBF during infusion of 50 ug/min of Acetylcholine minus resting FBF divided by resting FBF. Also, a comprehensive two-dimensional and Doppler echocardiography was performed with an Acuson XP124 cardiac ultrasound unit (Acuson, Mountain View, California, USA). LV dimensions were measured with M-mode. Left ventricular mass (LVM) was determined from the Penn convention and indexed for height to obtain left ventricular mass index (LVMI).<sup>18</sup>

In both ULSAM (fourth examination cycle) and PIVUS (second examination cycle), urine albumin was measured by nephelometry (Urine albumin, Dade Behring, Deerfield IL, USA) using a Behring BN ProSpec® analyzer (Dade Behring). Urine creatinine was analyzed with a modified kinetic Jaffe reaction on an Architect Ci8200® analyzer (Abbott, Abbot Park, IL, USA) and creatinine related urine albumin (ACR) was calculated.

### **Statistical analysis**

We initially investigated distributions of all variables. Serum endostatin was logarithmically transformed in all analyses in order to promote a normal distribution. We thereafter performed cohort-specific analyses in PIVUS and ULSAM of the association between the duration of

hypertension and serum endostatin (modelled as a continuous variable expressed per SD increase) using linear regression in the following multivariable models:

A) Age- and sex-adjusted;

B) Lifestyle and cardiovascular risk factor model (age, sex, diabetes, smoking, BMI, total cholesterol, HDL-cholesterol, lipid-lowering treatment, prevalent cardiovascular disease, level of physical activity).

In secondary analyses, we also added glomerular filtration rate to multivariable model B.

Missing values for covariates were estimated by multiple imputation.

Moreover, we investigated the association between self-reported duration of hypertension and endostatin levels in PIVUS. Also, the cross-sectional association between systolic and diastolic blood pressure per se and serum endostatin was investigated in both cohorts using crude models and models adjusted for specific antihypertensive medication.

As a second step, we investigated the cross-sectional association between endostatin and markers of hypertensive end-target organ damage (left ventricular mass, endothelial dependent vasodilation [PIVUS, examination cycle 1], and urinary albumin/creatinine [ULSAM and PIVUS (examination cycle 2)]) in participants with prevalent hypertension using multivariable linear regression (Model A and B, as described above with the addition of the duration of hypertension to multivariable model B). A two-sided p-value <0.05 was regarded as significant and Stata 12.1 (Stata Corp College Station, TX, USA) was used for all analyses.

## Results

Baseline characteristics of both cohorts are shown in Table 1.

### *Serum endostatin and the duration of hypertension*

In examination cycle 2 of the PIVUS cohort, participants with >5-years history of hypertension portrayed higher serum endostatin compared to normotensive participants, while no endostatin increase was seen in participants with new-onset hypertension (0- 5 years duration, Table 2) Associations were somewhat attenuated after adjustment for age, sex, established cardiovascular risk factors and glomerular filtration rate albeit still statistically significant (Models A-C, Table 2). Moreover, when using questionnaire-based data on the duration of hypertension (range of duration of hypertension 0-42 years, 1-year longer hypertension duration was associated with 0.01 SD, 95 % CI, 0.001-0.02,  $p=0.03$ , higher endostatin in multivariable model C. When we compared the increase in endostatin between examination cycle 1 and 2 in PIVUS in those with persistent hypertension at both examinations compared to those who remained normotensive, the increase in endostatin was 55 % higher in those with persistent hypertension vs. those that remained normotensive (mean increase in endostatin 14.0 ng/ml vs 9.0 ng/ml, respectively  $p=0.005$ ).

The association between the longer duration of hypertension and higher serum endostatin was replicated in the ULSAM cohort where participants with 27 years of hypertension had the highest levels of endostatin compared to normotensives (Table 2). There were no cross-sectional associations between systolic/diastolic blood pressure levels per se and serum endostatin in any cohort ( $p>0.29$  for all, data not shown).

*Serum endostatin and hypertensive end-target organ damage in participants with prevalent hypertension*

The mean  $\pm$  standard deviation of endostatin in participants with prevalent hypertension at the first examination cycle of PIVUS was 48 ng/ml  $\pm$  14 ng/ml and the mean endothelial dependent vasodilation and left ventricular mass was 514%  $\pm$  298% and 46 g/m<sup>2</sup>  $\pm$  13 g/m<sup>2</sup>, respectively. Higher circulating endostatin was associated with lower endothelial dysfunction and higher left ventricular mass in age- and sex-adjusted models (Model A, Table 3). This association was, however, abolished when taking cardiovascular risk factors and duration of hypertension into account (Table 3).

The mean  $\pm$  standard deviation of albumin/creatinine ratio in participants with prevalent hypertension in PIVUS (second examination cycle) and ULSAM was, 7  $\pm$  30 mg/ $\mu$ mol, 5  $\pm$  21 mg/ $\mu$ mol, respectively. Higher serum endostatin was significantly, associated with higher urinary albumin/creatinine-ratio in all multivariable models in both cohorts (Table 3).

As seen in Supplementary Table 1, Endostatin levels appears higher in all classes of antihypertensive drugs in both cohorts, which argues against an effect of any specific antihypertensive drug class as an explanation of our findings.

## Discussion

In two independent community-based cohorts of elderly, participants with long term hypertension portrayed elevated serum levels of endostatin independently of established cardiovascular risk factors and kidney function. In addition, cross-sectional associations between circulating endostatin, and vascular, myocardial and renal indices of hypertensive end target organ damage were found. To our knowledge, these associations have not been reported previously. Our data put forward serum endostatin as an interesting novel biomarker for hypertensive organ damage.

Circulating levels of endostatin has been suggested to be a marker of breakdown and remodeling of the extracellular matrix in various diseases, such as cancer or aortic aneurysms,<sup>19-22</sup> however whether this is true also for hypertension is less studied. It is well known that long term hypertension induces cardiovascular extra cellular matrix remodeling.<sup>4, 22</sup> Mechanical stretch of the vasculature, as seen in hypertension, induces vascular extra cellular remodeling by activation of MMP-2 and MMP-9,<sup>23</sup> which both are proteases that play an important role in the degradation of collagen XVIII to endostatin. Moreover, experimental studies show that damages to the vasculature,<sup>24</sup> the myocardium,<sup>25</sup> and the kidneys<sup>26</sup> lead to increased expression of endostatin in these tissues and that circulating concentrations of endostatin is elevated in patients with prevalent cardiovascular diseases<sup>27-31</sup> or chronic kidney disease.<sup>13</sup> Thus, the fact that endostatin was both associated with the duration of hypertension and indices of vascular, myocardial, and renal hypertensive target organ damage in the present study may indicate that higher circulating levels of endostatin mirrors an increased extra cellular remodeling that originates from these tissues. Still, based on the present observational study, no firm conclusions regarding from what tissue the elevated levels of endostatin originate from should be drawn, nor can we establish the underlying mechanism that leads to higher circulating endostatin levels. Also, in the present study it is not possible to

disentangle whether increased endostatin levels are a cause or a consequence of pathologic processes. To date, our understanding of a potential causal role for endostatin in these tissues is limited. Further mechanistic studies are needed to shed light on this issue.

In addition to the fact that endostatin is a break down product from collagen XVIII, endostatin is also a potent endogenous angiogenesis inhibitor and is regulated in balance with vascular endothelial growth factor (VEGF).<sup>9</sup> Endostatin has due to this role also found its place in the treatment of cancer, particularly small cell lung cancer.<sup>32</sup> VEGF levels are increased in hypertensive patients,<sup>33</sup> and a disturbed VEGF-mediated angiogenesis have been suggested to play a causal role in the development of vascular,<sup>34</sup> cardiac,<sup>35</sup> and renal<sup>36</sup> hypertensive damage. Thus, one possible explanation could be that increased endostatin mirrors a systemic elevated angiogenic activity reflecting increased neo-vascularisation induced by vascular, myocardial, or renal ischemia, similar as the role suggested for VEGF.<sup>4, 13, 14, 37-39</sup>

Endostatin has also been shown to exert acute reductions in blood pressure via a release of nitric oxide<sup>40</sup> and individuals with Down Syndrome have significantly higher circulating levels of endostatin and exhibit lower blood pressure compared with control subjects.<sup>41, 42</sup> This may indicate that endostatin, per se, may exert a blood pressure lowering effect. Thus speculatively, a possible explanation to our findings could be that endostatin is endogenously released in order to regulate the blood pressure as a response/protective mechanism to hemodynamic changes caused by remodeling of the extracellular matrix in the vascular tree or organs. However, the fact that no cross-sectional association was found between endostatin and blood pressure levels per se, would argue against a blood pressure regulating role of endostatin as an explanation of our findings.

Finally, it is also possible that other cardiovascular risk factors that are closely associated with both endostatin, long term hypertension and hypertensive target organ damage, such as age,<sup>43</sup> obesity,<sup>44, 45</sup> physical activity,<sup>43</sup> gluco-metabolic disturbance,<sup>44, 45</sup> and lipids,<sup>46</sup> may mediate the present associations. This appeared true for the association between endostatin and endothelial dysfunction and left ventricular mass but not for the association between endostatin and urinary albumin excretion rate or the duration of hypertension (Table 2 and 3). The association between the duration of hypertension and endostatin was further attenuated, albeit still statistically significant, after adjustments for kidney function in both cohorts suggesting that reduced renal clearance of serum endostatin can explain a portion of the present associations, or alternatively, that the reduced glomerular filtration rate is a yet another aspect of hypertensive renal damage that is reflected by endostatin levels.

Endostatin levels have been shown to be elevated in patients with various cardiovascular diseases,<sup>20, 27-29, 31, 47</sup> associations that also are supported by experimental data.<sup>46, 48, 49</sup> For example, higher endostatin levels has been shown to be both a predictor of ischemic strokes,<sup>27</sup> and with poorer outcomes in stroke patients.<sup>50</sup> Also in a recent study, we report that higher endostatin levels predict mortality from both cardiovascular disease and cancer in the present community based cohort.<sup>(Arnlov-ATVB-in press)</sup> Additional studies investigating are warranted to determine if endostatin levels can be considered a marker for subclinical cardiovascular damage used to identify hypertensive individuals at particularly increased cardiovascular risk.

One major strength of the present study is the replication of the results in an independent cohort. This approach increases the validity and generalizability of the results and limits the risk for reporting spurious associations. Also, with the use of two cohorts it was possible to take advantage of the unique strengths of each cohort such as longitudinal data with up to 27 years follow-up, or a detailed characterization of participants with regards to sub-clinical vascular, myocardial and renal organ damage.

Limitations include the limited generalizability to other age- and ethnic groups. Furthermore, as data on endostatin was only available at one examination cycle in ULSAM it was not possible to investigate the interplay between the hypertension and changes in endostatin levels over time in this cohort. Also, as current knowledge on factors that influence circulating levels of endostatin is very limited and we cannot rule out that cohort-specific effects due to differences in handling of the samples (such as freezer time), or differences in gender, age or the time of the baseline examination between the two cohorts may influence the absolute levels of endostatin. However, given the similarity of results between the two studies, this had most likely no major impact on our results.

### **Perspectives**

Our data indicate that circulating endostatin is associated with the duration of hypertension as well as with vascular, myocardial and renal indices of hypertensive target organ damage. Studies evaluating the underlying mechanisms and the clinical relevance of our findings are warranted.

### **Acknowledgments**

None.

### **Sources of Funding**

This study was supported by The Swedish Research Council (2006-6555, 2012-1727, 2012-2215), Swedish Heart-Lung foundation, Thuréus foundation, the Marianne and Marcus Wallenberg Foundation, Dalarna University and Uppsala University

### **Conflict of interest/disclosures**

There are no conflicts of interest. The funding sources did not play any role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; and

preparation, review, or approval of the manuscript. Dr. Ärnlov is the guarantor of this work, had full access to all the data, and takes full responsibility for the integrity of data and the accuracy of data analysis.

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## **Novelty and significance**

### **What is new?**

An association between longer duration of hypertension and serum levels of endostatin was found and validated in 2 independent cohorts. Participants with more than 27 years history of hypertension had the highest endostatin levels. Interestingly, in participants with prevalent hypertension, we found cross-sectional associations between higher circulating endostatin and impaired endothelial function, increased left ventricular mass and higher urinary albumin/creatinine ratio.

### **What is relevant?**

Endostatin, a biologically active derivate of collagen XVIII, has been suggested to be a relevant marker for extracellular matrix turnover and remodeling in various disease. Yet, the role of endostatin in hypertension and hypertensive target organ damage is unclear.

### **Summary**

Our data provide additional support for the importance of increased extracellular remodeling in hypertensive disease and put forward serum endostatin as a novel biomarker for hypertensive organ damage.

### **Figure legends**

There are no figures.

**Table 1.** Baseline characteristics in the PIVUS and ULSAM cohorts

<b>Variable</b>	<b>PIVUS</b>	<b>ULSAM</b>
Number of subjects	812	785
Female no. (%)	414 (51)	0 (0)
Age (years)	75.3 ±0.2	77.6 ±0.8
Body mass index (kg/m <sup>2</sup> )	26.8 ±4.3	26.3 ±3.5
Serum total cholesterol (mg/dL)	5.4 ±1.1	5.4 ±1.0
Serum HDL cholesterol (mg/dL)	1.5 ±0.5	1.3 ±0.3
Fasting plasma glucose mg/dL)	5.2 ±1.5	5.9 ±1.4
Systolic blood pressure (mmHg)	149 ±19	151 ±21
Serum endostatin (ng/ml)	60 ±27	55 ±18
Glomerular filtration rate	68 ± 19	73 ± 17
Smoking – no. (%)	50 (6)	59 (8)
Diabetes no. (%)	112 (14)	112 (14)
Previous cardiovascular disease- no. (%)	164 (20)	213 (27)
Lipid lowering treatment- no. (%)	216 (27)	131 (17)
Beta-blockers	231 (28)	195 (25)
Ca-antagonists	133 (16)	121 (15)
Diuretics	181 (22)	130 (17)
RAAS-blockade (ACE and ARB)	251 (31)	132 (17)
Leisure time physical activity- no. (%)		
-Sedentary	99 (12)	62 (8)
-Moderate	498 (61)	274 (35)
-Regular	172 (21)	413 (53)
-Athletic	43 (5)	36 (5)

Data are mean ± standard deviation for continuous variables and n (%) for categorical variables

**Table 2** The association between the duration of exposure to hypertension and endostatin in the PIVUS and ULSAM cohorts: Multivariable linear regression

<b>PIVUS cohort</b>		<b>Model A</b>			<b>Model B</b>		<b>Model C</b>	
<b>Time of exposure</b>	<b>N (no,%)</b>	<b>Mean endostatin (ng/ml) ± standard deviation</b>	<b>B coefficient (95% CI)</b>	<b>p</b>	<b>B coefficient (95% CI)</b>	<b>p</b>	<b>B coefficient (95% CI)</b>	<b>p</b>
No hypertension at baseline	124 (135)	53±13	Referent	Referent	Referent	Referent	Referent	Referent
0-5years duration of hypertension	103 (13)	55±14	0.12 (-0.14-0.38)	0.35	0.08 (-0.17-0.34)	0.52	0.08 (-0.15-0.30)	0.50
>5 years history of hypertension	585 (72)	62±30	0.42 (0.23-0.61)	<0.001	0.33 (0.13-0.53)	0.001	0.25 (0.08-0.42)	0.005
<b>ULSAM cohort</b>								
No hypertension at baseline	96 (12)	48±12	Referent	Referent	Referent	Referent	Referent	Referent

0-6 years duration of hypertension	97 (12)	56±24	0.45 (0.18-0.73)	0.001	0.39 (0.12-0.67)	0.005	0.20 (-0.03-0.43)	0.09
7-16 years duration of hypertension	181 (23)	54±18	0.33 (0.084-0.57)	0.008	0.27 (0.03-0.51)	0.026	0.17 (-0.03-0.38)	0.10
17-27 years duration of hypertension	134 (17)	57±15	0.55 (0.29-0.81)	<0.001	0.45 (0.20-0.71)	0.001	0.31 (0.09-0.53)	0.005
>27 years duration of hypertension	277 (35)	57 ±17	0.57 (0.35-0.80)	<0.001	0.47 (0.24-0.70)	<0.001	0.30 (0.11-0.50)	0.003

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Regression ( $\beta$ ) are expressed as SD increase of serum endostatin. Model A) Age-adjusted; Model B) Lifestyle and cardiovascular risk factor model (age, diabetes, smoking, BMI, total cholesterol, HDL-cholesterol, lipid-lowering treatment, prevalent cardiovascular disease, level of physical activity). Model C) All variables in model B and also glomerular filtration rate.

**Table 3** The association between endostatin and different indices of hypertensive target organ damage (endothelial function, left ventricular mass and urinary albumin creatinine ratio) in participants with prevalent hypertension in the PIVUS and ULSAM cohorts: Multivariable linear regression

<b>End organ damage</b>		<b>Model A</b>		<b>Model B</b>	
<b>PIVUS cohort</b>	<b>Number of participants</b>	<b>B coefficient (95% CI)</b>	<b>p</b>	<b>B coefficient (95% CI)</b>	<b>p</b>
Endothelial function*	611	- 0.09 (-0.16 - -0.01)	P=0.03	-0.32 (-0.11 – 0.05)	P=0.42
Left ventricular mass*	647	0.16 (0.08 – 0.23)	P < 0.0001	0.06 (-0.02 – 0.15)	P=0.144
Urinary albumin/creatinine-ratio	560	0.16 (0.08 – 0.24)	P < 0.0001	0.13 (0.05 – 0.22)	P = 0.0002
<b>ULSAM cohort</b>		<b>B coefficient (95% CI)</b>	<b>p</b>	<b>B coefficient (95% CI)</b>	<b>p</b>
Urinary albumin/creatinine-ratio	531	0.22 (0.14 – 0.30)	P < 0.0001	0.21 (0.13 – 0.29)	P < 0.0001

Regression ( $\beta$ ) are expressed as SD increase of serum endostatin per SD increase of the different indices of hypertensive target organ damage Model A) Age and sex-adjusted; Model B) Model A and cardiovascular risk factor model (age, diabetes, smoking, BMI, total cholesterol, HDL-cholesterol, lipid-lowering treatment, prevalent cardiovascular disease, level of physical activity and the duration of hypertension).

\* Performed at the first examination cycle of PIVUS.

