FGF23 and atherosclerosis

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Relationship between circulating FGF23 and total body atherosclerosis in the community

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Abstract
Background. Fibroblast growth factor-23 (FGF23) is a regulator of mineral metabolism and has been suggested to play a role in vascular calcification in chronic kidney disease (CKD). Data on the association between FGF23 and atherosclerosis, both in CKD and in the community, is limited.

Methods. The total body atherosclerosis score (AS) was determined by a magnetic resonance imaging-based angiography in 306 elderly men and women, representing a subsample of the community-based PIVUS cohort. Subjects were divided into three categories based on AS: AS = 0, low AS and high AS. Serum FGF23 was measured using a two-site monoclonal antibody ELISA.

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Introduction

Fibroblast growth factor-23 (FGF23) is a phosphaturic hormone that was initially cloned as the causative factor for the hereditary disorder autosomal dominant hypophosphataemic rickets (ADHR) [1]. FGF23 is produced in bone and regulates serum phosphate through decreasing the reabsorption of phosphate in the kidney proximal tubule [2–4]. FGF23 also controls vitamin D metabolism through decreasing the renal expression of 1-alpha-hydroxylase and increasing that of 24-hydroxylase [5,6].

Serum FGF23 is increased in parallel with a decline in renal function [7,8]. Elevated serum FGF23 levels have been linked to the development of secondary hyperparathyroidism [9] and predict the progression of renal failure [10] as well as total mortality in incident haemodialysis patients [11]. Although atherosclerosis and vascular dysfunction are main contributors to the high prevalence of cardiovascular morbidity and mortality in patients with chronic kidney disease (CKD) [12,13], data on the association between circulating FGF23 and atherosclerosis, both in CKD and in the general population, is limited.

With the evolving concept of whole-body magnetic resonance angiography (WBMRA) [14], it is possible to assess the whole arterial tree in one session without ionizing radiation. This minimally invasive global atherosclerotic assessment enables studies of the relationship between the burdens of vascular stenosis and occlusions and various constitutional and metabolic cardiovascular risk factors. It has previously been shown, using digital subtraction angiography as the gold standard, that WBMRA has a high sensitivity and specificity for estimating the extent of atherosclerosis in patients with peripheral vascular disease [15–17]. Furthermore, the atherosclerosis score (AS) is associated with several cardiovascular risk factors in the normal population [18,19]. The value of WBMRA both in clinical practice and in patient-based studies is yet to be established.

One of several aims of the Prospective Investigation of the Vasculature in Uppsala Seniors (PIVUS study) is to evaluate the predictive power of multiple variables for the development of atherosclerosis in elderly subjects [20]. Herein, we analysed the relationship between FGF23 and atherosclerosis in 306 individuals (representing a subsample of the PIVUS cohort) who had undergone WBMRA measurements. All analyses were defined a priori with pre-specified subgroup analysis in subjects without previous cardiovascular disease.

Subjects and methods

Study participants

The PIVUS study includes 1016 men and women aged 70 of which 306 (145 women, 161 men) underwent WBMRA in a clinical 1.5-T scanner and were included in our primary analyses. Secondary analyses were also performed in participants without previous cardiovascular disease (n = 261). Participants in the PIVUS cohort were chosen from the population register of the community and were invited within 2 months of their 70th birthday in a randomized order from April 2001 to June 2004. No significant differences in the basic characteristics and major cardiovascular risk factors were found between the total PIVUS sample, the WBMRA subsample and those not attending the basic investigation [18,20]. The ethics committee of Uppsala University approved the study and the participants gave written informed consent.

Basic investigation

Plasma cystatin C (reagent: 1014, Gentian, Moss, Norway), creatinine (reagent: 14.3600.01, Synermed International, Westfield, IN, USA), phosphate (reagent: 7D71-30) and urea (reagent: 7D75-20) measurements were performed on an Architect Ci8200 analyser (Abbott Laboratories, Abbott Park, IL, USA) and reported using SI units. Estimated glomerular filtration rate (eGFR) was derived from cystatin C, by the formula eGFRcystC = 79 901 × CystC−1.4389. The assay has high precision, good linearity and strong correlation with iohexol clearance (R² = 0.956) [21]. Impaired renal function was defined as eGFR < 60 mL/min/1.73 m². The LIAISON 25(OH)D3 assay (DiaSorin Inc., Saluggia, Italy) was performed on the LIAISON analyser according to the manufacturer’s instructions. PTH levels were analysed using the Immulite 2000 intact PTH assay (Diagnostic Products Corporation, Los Angeles, CA, USA). Lipid variables, calcium and fasting blood glucose were measured by standard laboratory techniques at Uppsala University Hospital. Smoking status, current smoking versus nonsmoking, was obtained from a questionnaire. Previous cardiovascular disease was defined as a history of any of the following events: myocardial infarction, stroke, heart failure, angina pectoris or surgical procedures for angina/severe coronary disease. All subjects were investigated under standardized conditions in the morning after an overnight fast. No medication or smoking was allowed after midnight.

Serum FGF23

Intact FGF23 was measured using an ELISA according to the manufacturer’s protocol (Kainos Laboratories International; Tokyo, Japan) [22]. This second-generation, two-site, monoclonal antibody ELISA has previously been shown to recognize the biologically active, intact FGF23 [22]. The Kainos intact FGF23 assay has a lower limit of detection of 3 pg/mL and intra- and interassay coefficients of variation of less than 5%. The Kainos intact assay was the most sensitive among three different two-site enzyme-linked immunosorbent assays for FGF23 measurements [23].

Whole body magnetic resonance angiography (WBMRA)

The WBMRA method has previously been described in detail [18,19]. In brief, a gadolinium contrast agent was administered intravenously and the WBMRA examination was divided into four stations. The first station included the supra-aortic arteries and the thoracic aorta. The second station

Results. In continuous and multi-category regression models, higher FGF23 was associated with a significant increase in the odds of having a high AS (OR 1.43, CI 1.06–1.92 to OR 3.01, CI 1.52–5.99). This association was stronger in individuals with eGFR < 60 mL/min/1.73 m² (n = 27), reaching a nearly 6-fold increase in the odds for a high AS in the upper FGF23 tertile (OR 5.64, CI 2.78–11.5). We found weaker support for a relationship between FGF23 and the presence of atherosclerosis as subjects in the highest FGF23 tertile had an increased risk for an AS > 0 in crude models (OR 1.93, CI 1.05–3.55), but this was not statistically significant in adjusted (OR 1.42, CI 0.74–1.72) models.

Conclusions. We provide novel evidence supporting an association between serum FGF23 and total body atherosclerosis in the community. Additional studies are warranted to determine the prospective relationship between FGF23 and atherosclerosis, and whether FGF23 is a modifiable cardiovascular risk factor.

Keywords: atherosclerosis; calcification; chronic kidney disease; FGF23; phosphate
contained the abdominal aorta, including the renal arteries and the third station started at the external iliac arteries and continued to the popliteal arteries. The fourth and last station continued for a varying distance below the ankle. An overlap of 3 cm between each station gave a maximum total length of coverage of 171 cm. Vessels that were overlapped were assessed on both stations, and there was no difference when assessing the degree of stenosis in a vessel that was overlapped. The resulting images were manually evaluated and the degree of stenosis was measured with callipers on a workstation. The AS was defined as the sum of vascular abnormalities for each of the five different vascular territories; neck, aorta, kidney, upper leg and lower leg, was assessed. A normal vessel segment received null points, <50% stenosis one point, and ≥50% stenosis and occlusion received two points. The sum for each territory was then divided by the maximum achievable sum for that territory, and the ratio was multiplied by 100. For each segment, only the most severe stenosis or occlusion was noted. The total body AS is the sum of the AS for all five vascular territories divided by five. Approximately one-third of the sample had no vascular abnormalities, one-third had stenosis of <50% and the remainder had stenosis ≥50% or occlusions [18]. The intra-observer reproducibility was good (kappa value = 0.73) with intra-observer agreement in 94% of the segments. Inter-observer reproducibility was excellent (kappa value = 0.83) with inter-observer agreement in 77% of the segments [18]. The mean length of time between the basic PIVUS investigation and WBMRA was 16 months (range 3–24 months). Exclusion criteria for the WBMRA examination were pacemaker, valvular prostheses, intra-cranial clips and claustrophobia.

Statistical analyses
Logarithmic transformation was performed to achieve normal distribution of skewed variables (FGF23, calcium and PTH).

In our primary analysis, we modelled FGF23 as a linear continuous variable [expressed as a one standard deviation (1-SD) increase]. We also used multi-category and threshold models comparing the odds in FGF23 tertiles 2 and 3 to those in tertile 1 (lowest) and the odds in tertile 3 to those in tertiles 1 and 2 combined. Thresholds were based on the FGF23 distribution in our sample. Since the AS variable did not follow the Poisson distribution, which count variables often do, it was further divided into categorical variables used in all subsequent analyses: (1) no stenosis (AS = 0): 100 subjects; (2) low AS (AS = 0–7): 99 subjects; (3) high AS (AS ≥ 7): 107 subjects. We addressed the following questions: are increased FGF23 levels associated with higher odds of having arterial stenosis (AS > 0 compared to AS = 0), and are increased FGF23 levels associated with increased odds of a high AS compared to low AS in subjects with AS > 0 (n = 206).

Three logistic regression models were used for all analyses: model A, crude model; model B, adjusted for gender and biochemical factors (phosphate, calcium, PTH, 25(OH)D3 and eGFR) and model C, adjusted for the Framingham risk score (a compound cardiovascular risk score) [24]. Finally, we investigated whether the known interaction between FGF23 and renal function [8,25–27] alters the association between FGF23 and total body AS, by invoking a multiplication term between FGF23 tertiles and the presence of impaired renal function (eGFR < 60 mL/min/1.73 m²) in the regression models. P-values < 0.05 from two-sided tests were considered significant. The statistic software package STATA 10.1 (Stata Corporation, College Station, USA) was used for all calculations.

Results
Clinical characteristics, including cardiovascular risk factors, in the whole sample, over AS categories and over FGF23 tertiles are presented in Table 1. The median serum FGF23 level in the whole sample was 43.1 pg/mL, ranging from 15.3 to 192 pg/mL. A total of 100 subjects had a total body AS equal to 0 (FGF23 median 41.3 pg/mL and IQR 34.4–49.2 pg/mL), and 206 subjects had total body AS ranging from 1 to 27.5 and a median of 7. We used the median value as the cut-off between subjects with a low AS (99 subjects; FGF23 median 40.4 pg/mL and IQR 31.5–52.5 pg/mL) and subjects with a high AS (107 subjects; FGF23 median 48.7 pg/mL and IQR 38.1–59.0 pg/mL). FGF23 levels were found to be significantly increased in the high AS group compared to the low AS group (P < 0.001).

FGF23 as a marker of the severity of stenosis (high versus low AS)

The 1-SD increase of FGF23 was associated with a 43–49% increased odds of having a high total body AS versus a low total body AS in both crude and adjusted models (Table 2). These results were confirmed in both multi-category and threshold models (Table 2). Subjects in the highest FGF23 tertile (> 50.2 pg/mL) had nearly a 3-fold increase risk of a high AS compared to the lowest tertile. The proportions of individuals who had a high AS over FGF23 tertiles are shown in Figure 1A, supporting a graded relationship between FGF23 and degree of atherosclerosis. Importantly, adjustment for serum phosphate did not attenuate our results (model B), and we found no correlation between phosphate and AS (data not shown). Secondary analyses in subjects without previous cardiovascular disease showed similar results with somewhat lower odds ratios (Table 2). To exclude the possibility that the associations were driven by extreme FGF23 values, we excluded subjects with FGF23 levels not within two SDs from the mean (n = 7, range 93.6–192 pg/mL). Importantly, removal of those subjects revealed a stronger association between FGF23 and the severity of stenosis (OR 1.82 CI 1.29–2.57 for 1-SD increase in log FGF23).

FGF23 as a marker for the presence of stenosis

We found evidence for a non-linear relationship between FGF23 and the presence of stenosis, with an increased risk for a positive AS in the highest FGF23 tertile (OR 1.73, CI 1.05–3.55) in crude models, although nonsignificant in multivariate adjusted model B (OR 1.42, CI 0.74–1.72). The non-linear relationship between FGF23 and a positive AS was confirmed graphically in Figure 1B, showing the proportion of individuals over FGF23 tertiles who had an AS > 0. The relationship between FGF23 and the presence of stenosis was nonsignificant in continuous models or subgroup analyses in subjects without previous cardiovascular disease (Table 2). Results were similar when excluding subjects with FGF23 levels not within two SDs from the mean (data not shown).

Interaction between FGF23 and renal function

A total of 27 subjects had an age-adjusted diminished renal function (eGFR < 60 mL/min/1.73 m², mean eGFR: 51.5 ± 8.0 mL/min/1.73 m²). The odds ratios for FGF23 predicting the degree of AS were significantly higher in subjects with eGFR < 60 mL/min/1.73 m² (crude model), supporting a nearly 6-fold increased risk for a high AS in the upper FGF23 tertile (Table 3). We found no evidence for the interaction term when predicting the presence of stenosis. Collectively, the increased risk for a high AS followed by...
Table 1. Basic characteristics of the study population

<table>
<thead>
<tr>
<th>Whole WBMRAsample</th>
<th>AS = 0</th>
<th>Low AS</th>
<th>High AS</th>
<th>FGF23 tertile1 (&lt;37.4 pg/mL)</th>
<th>FGF23 tertile2 (37.4–50.2 pg/mL)</th>
<th>FGF23 tertile3 (&gt;50.2 pg/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of subjects (n)</td>
<td>306</td>
<td>100</td>
<td>99</td>
<td>107</td>
<td>102</td>
<td>102</td>
</tr>
<tr>
<td>Females</td>
<td>145 (47.4)</td>
<td>60 (60)</td>
<td>54 (54.6)</td>
<td>31 (29.0)</td>
<td>52 (51.0)</td>
<td>53 (52.0)</td>
</tr>
<tr>
<td>Basic characteristics and biochemical analyses</td>
<td></td>
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<tr>
<td>Height (cm)</td>
<td>169 ± 9.4</td>
<td>167 ± 9.2</td>
<td>169 ± 8.7</td>
<td>172 ± 9.5</td>
<td>169 ± 9.5</td>
<td>169 ± 9.2</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>77 ± 14.1</td>
<td>75 ± 12.4</td>
<td>76 ± 14.2</td>
<td>80.5 ± 15.0</td>
<td>76 ± 15.2</td>
<td>77.2 ± 13.3</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>26.9 ± 4.1</td>
<td>26.9 ± 3.7</td>
<td>26.6 ± 4.2</td>
<td>27.1 ± 4.3</td>
<td>26.6 ± 4.6</td>
<td>27.0 ± 4.0</td>
</tr>
<tr>
<td>Systolic blood pressure (mmHg)</td>
<td>149 ± 22.3</td>
<td>146 ± 19.6</td>
<td>147 ± 21.3</td>
<td>154 ± 24.8</td>
<td>147 ± 20.0</td>
<td>151 ± 21.1</td>
</tr>
<tr>
<td>Diastolic blood pressure (mmHg)</td>
<td>78 ± 10.0</td>
<td>78 ± 9.1</td>
<td>76 ± 9.9</td>
<td>80 ± 10.6</td>
<td>78 ± 10.8</td>
<td>78 ± 9.7</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>61 ± 8.7</td>
<td>62 ± 9.4</td>
<td>62 ± 8.5</td>
<td>60 ± 8.1</td>
<td>61 ± 8.3</td>
<td>61 ± 9.2</td>
</tr>
<tr>
<td>Serum cholesterol (mmol/L)</td>
<td>5.4 ± 1.0</td>
<td>5.4 ± 0.9</td>
<td>5.4 ± 1.1</td>
<td>5.4 ± 0.9</td>
<td>5.4 ± 0.9</td>
<td>5.4 ± 1.0</td>
</tr>
<tr>
<td>Serum low-density lipoprotein (mmol/L)</td>
<td>3.3 ± 0.8</td>
<td>3.3 ± 0.8</td>
<td>3.3 ± 0.9</td>
<td>3.4 ± 0.8</td>
<td>3.3 ± 0.8</td>
<td>3.4 ± 0.9</td>
</tr>
<tr>
<td>Serum triglycerides (mmol/L)</td>
<td>1.3 ± 0.6</td>
<td>1.2 ± 0.7</td>
<td>1.3 ± 0.6</td>
<td>1.4 ± 0.6</td>
<td>1.3 ± 0.7</td>
<td>1.3 ± 0.5</td>
</tr>
<tr>
<td>Serum glucose (mmol/L)</td>
<td>5.3 ± 1.6</td>
<td>5.2 ± 1.2</td>
<td>5.3 ± 1.6</td>
<td>5.5 ± 1.9</td>
<td>5.0 ± 0.9</td>
<td>5.4 ± 1.0</td>
</tr>
<tr>
<td>Serum phosphate (mmol/L)</td>
<td>1.1 ± 0.2</td>
<td>1.1 ± 0.2</td>
<td>1.1 ± 0.2</td>
<td>1.1 ± 0.2</td>
<td>1.1 ± 0.2</td>
<td>1.1 ± 0.2</td>
</tr>
<tr>
<td>Serum calcium (mmol/L)</td>
<td>2.3 (2.2–2.5)</td>
<td>2.3 (2.2–2.5)</td>
<td>2.3 (2.2–2.5)</td>
<td>2.4 (2.2–2.5)</td>
<td>2.3 (2.2–2.5)</td>
<td>2.4 (2.2–2.5)</td>
</tr>
<tr>
<td>Serum 25(OH)D (nmol/L)</td>
<td>61.1 ± 20.3</td>
<td>59.6 ± 21.1</td>
<td>63.3 ± 19.0</td>
<td>60.6 ± 20.7</td>
<td>60.1 ± 19.9</td>
<td>60.3 ± 21.4</td>
</tr>
<tr>
<td>Estimated glomerular filtration rate (mL/min/1.73 m²)</td>
<td>43.0 (25.0–68.8)</td>
<td>41.9 (22.0–67.5)</td>
<td>41.2 (23.4–65.9)</td>
<td>45.2 (26.7–78.9)</td>
<td>43.3 (24.6–67.7)</td>
<td>41.6 (25.3–67.2)</td>
</tr>
<tr>
<td>Fibroblast growth factor-23 (pg/mL)</td>
<td>43.0 (27.4–67.9)</td>
<td>41.3 (27.4–66.8)</td>
<td>40.35 (25.4–67.9)</td>
<td>48.7 (29.7–70.1)</td>
<td>40.6 (23.4–36.2)</td>
<td>30.6 (23.4–49.0)</td>
</tr>
<tr>
<td>Framingham risk score</td>
<td>11.0 ± 3.3</td>
<td>10.5 ± 3.2</td>
<td>10.7 ± 3.3</td>
<td>12.2 ± 3.2</td>
<td>10.6 ± 3.2</td>
<td>11.5 ± 3.5</td>
</tr>
<tr>
<td>Smoking habits and previous cardiovascular disease status</td>
<td></td>
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<tr>
<td>Smoker</td>
<td>24 (7.8)</td>
<td>3 (3)</td>
<td>5 (5.1)</td>
<td>16 (15.0)</td>
<td>7 (6.9)</td>
<td>8 (7.8)</td>
</tr>
<tr>
<td>Previous cardiovascular disease</td>
<td>45 (14.7)</td>
<td>7 (7)</td>
<td>12 (12.1)</td>
<td>26 (24.3)</td>
<td>9 (8.8)</td>
<td>13 (12.8)</td>
</tr>
</tbody>
</table>

Values are mean ± SD for normally distributed continuous variables, median (10th–90th percentiles) for non-normally distributed variables and n (%) for categorical variables.
Table 2. Relationship between FGF23 and atherosclerosis score (AS)

<table>
<thead>
<tr>
<th></th>
<th>FGF23 as a marker for the severity of stenosis (high versus low total body AS)</th>
<th>FGF23 as a marker for the presence of stenosis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Model A</td>
<td>Model B</td>
</tr>
<tr>
<td>Whole cohort</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(n = 306)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Continuous models</td>
<td>1-SD increase</td>
<td>Multi-category models</td>
</tr>
<tr>
<td>Tertile 1 (&lt;37.4 pg/mL)</td>
<td>Referent</td>
<td>Referent</td>
</tr>
<tr>
<td>Tertile 2 (37.4–50.2 pg/mL)</td>
<td>Referent</td>
<td>Referent</td>
</tr>
<tr>
<td>Tertile 3 (&gt;50.2 pg/mL)</td>
<td>Referent</td>
<td>Referent</td>
</tr>
<tr>
<td>Threshold models</td>
<td>T1 versus T2</td>
<td>T1 versus T2</td>
</tr>
<tr>
<td>Subjects without previous cardiovascular disease (n = 261)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Continuous models</td>
<td>1-SD increase</td>
<td>Multi-category models</td>
</tr>
<tr>
<td>Tertile 1 (&lt;37.4 pg/mL)</td>
<td>Referent</td>
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<tr>
<td>Tertile 2 (37.4–50.2 pg/mL)</td>
<td>Referent</td>
<td>Referent</td>
</tr>
<tr>
<td>Tertile 3 (&gt;50.2 pg/mL)</td>
<td>Referent</td>
<td>Referent</td>
</tr>
<tr>
<td>Threshold models</td>
<td>T1 versus T2</td>
<td>T1 versus T2</td>
</tr>
</tbody>
</table>

Model A: crude model. Model B: adjusted for serum phosphate, calcium, 25(OH)D3, PTH, eGFR and gender. Model C: adjusted for the Framingham risk score. Values are odds ratios (95% CI).

*P < 0.05, **P < 0.01.

Significant values are shown in bold.

Fig. 1. (A) Proportion of subjects with a high atherosclerosis score (AS) over FGF23 tertiles for participants with AS > 0 [n = 206, chi2 (trend) P = 0.002]. (B) Proportion of subjects with a positive AS over FGF23 tertiles in the whole population [n = 306, chi2 (trend) P = 0.036]. Values are percent + 95% CI.

Table 3. Interaction between FGF23 and renal function on the impact on severity of atherosclerosis score (AS)

<table>
<thead>
<tr>
<th></th>
<th>All subjects with AS &gt; 0</th>
<th>Subjects with AS &gt;0 and eGFR ≥60 mL/min/1.73 m²</th>
<th>Subjects with AS &gt;0 and eGFR &lt;60 mL/min/1.73 m²</th>
</tr>
</thead>
<tbody>
<tr>
<td>FGF23 tertile 1</td>
<td>Referent</td>
<td>Referent</td>
<td>Referent</td>
</tr>
<tr>
<td>FGF23 tertile 2</td>
<td>2.15 (1.06–4.37)^*</td>
<td>1.79 (0.86–3.71)</td>
<td>2.42 (1.02–5.73)^**</td>
</tr>
<tr>
<td>FGF23 tertile 3</td>
<td>3.01 (1.52–5.99)^**</td>
<td>2.35 (1.56–4.77)^*</td>
<td>5.64 (2.78–11.5)^**</td>
</tr>
</tbody>
</table>

Note the increased risk for a high AS with increasing FGF23 levels in subjects with eGFR <60 mL/min/1.73 m² compared to those with eGFR ≥60 mL/min/1.73 m². Values are odds ratios (95% CI) adjusted for the interaction between FGF23 and renal function.

*P < 0.05, **P < 0.01.

Significant values are shown in bold.
higher FGF23 levels is further pronounced when eGFR falls below 60 mL/min/1.73 m².

Discussion

In the current study, we for the first time report an association between circulating FGF23 and total body atherosclerosis in vivo. We found evidence for a graded relationship between FGF23 and severity of atherosclerosis, whereas only subjects in the upper FGF23 tertile were at increased risk for having a positive versus a non-detectable AS measured by the WBMRA technique. It should be pointed out that all measurements were performed in a community-based setting with serum FGF23 and other biochemistries within the normal range. The data are consistent with our recent findings that higher FGF23 associates with endothelium dysfunction [28], which has been shown to precede development of atherosclerosis and also predict future cardiovascular outcomes [29–32].

To our knowledge, no previous investigations have explored the relationship between FGF23 and generalized atherosclerosis in the community. One study reported that FGF23 did not correlate with coronary artery calcification in patients with normal renal function [33]. Another study by Inaba et al. showed that FGF23 is linked to peripheral vascular calcification, but in prevalent haemodialysis patients [34].

Our results may be relevant in CKD, since the subgroup of individuals with eGFR < 60 mL/min/1.73 m² suffered from nearly a 6-fold increased odds of having a high AS when in the highest FGF23 tertile compared to the lowest. In this regard, the benefit of FGF23 measurement in CKD has been proposed as it predicts faster progression of renal disease in non-diabetic CKD patients [10] as well as mortality in incident haemodialysis patients [11]. Additional evidence for the use of FGF23 in determination of clinical outcomes in CKD is derived from Nakanishi and colleagues [35], who reported that FGF23 is a predictor of secondary hyperparathyroidism. The use of FGF23 as a clinical marker or for predicting cardiovascular outcomes remains to be established.

The positive association between FGF23 and atherosclerosis is in concert with the fact that higher FGF23 is related to worse outcome in CKD. However, it should be pointed out that FGF23 decreases serum phosphate and lower phosphate levels are inevitably linked to a reduction, not an increase, in cardiovascular risk in hemodialysis patients [36]. Therefore, it is possible that increased FGF23 may be a response to, rather than a cause of, atherosclerosis. This in analogy with the positive relationship between FGF23 and serum phosphate observed in CKD due to a compensatory increase in FGF23 related to hyperphosphataemia [10]. It will be of importance to investigate whether FGF23, like many other bone-related proteins, is expressed locally in the vessel wall during the process of atherosclerotic plaque formation [37].

It remains to be determined whether FGF23 plays a direct role in vascular pathology, or if FGF23 modulates atherosclerotic development indirectly through regulation of mineral metabolism. For example, animal studies support that Fgf23 null mice display extensive vascular calcification and nephrosclerosis [4]; however, Stubbs and co-workers showed that these vascular abnormalities were largely normalized at a low-phosphorous diet [38]. Thus, FGF23 regulation of serum phosphate appears to be an important mechanism to modulate vascular calcification. Regardless, our findings provide evidence for an independent relationship between FGF23 and atherosclerosis in the general population, based on that (1) we investigated a community-based setting without apparent abnormalities in mineral metabolism; (2) the results remained unaltered in subjects with normal FGF23 levels and after adjustment for biochemical covariates; and (3) we found no correlation between serum phosphate and atherosclerosis.

This study provides several strengths. The cohort size is large, and the phenotypic characterization with the WBMRA technique is novel. Our study is community based with inclusion of both genders. Finally, the results are not confounded by concomitant renal disease. Some potential limitations should also be acknowledged. We cannot rule out the possibility of residual confounding, e.g. we did not adjust for unmeasured or yet unknown factors, such as bone status, 1,25(OH)₂D₃ levels or genetic variations contributing to atherosclerosis. We did not determine urinary phosphate excretion, a more accurate marker of phosphate load than serum phosphate in subjects with normal renal function. Participants were elderly Caucasian subjects, which may limit the generalizability of the results. The formulation of the AS variable may be a limitation, especially since there could be significant heterogeneity among individuals with the same score, but also because almost one-third of the subjects had an AS of zero.

In conclusion, we report on a novel association between FGF23 and atherosclerosis in the community. Additional studies are warranted to evaluate if FGF23 can predict cardiovascular outcomes, and whether it is a modifiable risk factor.

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References


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