Editorial Review

The role of FGF-23 in CKD–MBD and cardiovascular disease:
friend or foe?

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Introduction

Regulation of mineral metabolism is a complex process, and our current models are constantly revised. The cloning and characterization of fibroblast growth factor-23 (FGF-23) has revolutionized our understanding of the endocrine regulation underlying phosphate and vitamin D metabolism. FGF-23 was initially regarded as a pathogenic factor primarily involved in hereditary skeletal disorders of hypophosphataemic rickets [1–3], however the area of research related to FGF-23 has significantly expanded into its present role as a key player in chronic kidney disease (CKD–MBD). From a clinical perspective, associations between circulating FGF-23, cardiovascular risk factors and mortality should be recognized. Although causal relationships between excess FGF-23 and adverse outcomes remain to be proven, this review attempts to identify potential mechanisms that presumably link FGF-23 to an increased cardiovascular risk.

Physiology and function of FGF-23

FGF-23 was initially identified as the causative factor of autosomal dominant hypophosphataemic rickets, characterized by hypophosphataemia due to reduced renal reabsorption of phosphate, inappropriately low to normal 1,25-dihydroxyvitamin D3 (1,25(OH)2D3) levels and rickets/osteomalacia [1]. FGF-23 was also identified as an ectopically overproduced phosphaturic factor in tumour-induced osteomalacia [4]. Collectively, FGF-23 emerged as a novel negative regulator of circulating phosphate and 1,25(OH)2D3 levels, subsequently confirmed in extensive animal and in vitro studies. FGF-23 induces phosphaturia through decreased expression and endocytosis of the sodium–phosphate co-transporters Npt2a and Npt2c in the kidney proximal tubule, and directly suppresses renal 1-alpha-hydroxylase, leading to decreased conversion of 25-hydroxyvitamin D3 (25(OH)D3) to its active metabolite 1,25(OH)2D3 [5–11]. Another mechanism by which FGF-23 reduces both 25(OH)D3 and 1,25(OH)2D3 levels is stimulation of the 24-hydroxylase, since this enzyme has the capacity to inactivate these metabolites to the vitamin D degradation pathway [12].

The physiological actions of FGF-23 in phosphate and vitamin D metabolism are evidenced by several hereditary syndromes, leading either to excess or deficiency of circulating, biologically active FGF-23. FGF-23 excess is observed in autosomal dominant hypophosphataemic rickets due to activating mutations in the FGF-23 gene [1], causing FGF-23 resistance to proteolytic degradation [13]. Similarly, inactivating mutations in the PHEX or DMP-1 genes lead to X-linked hypophosphataemic rickets and autosomal recessive hypophosphataemic rickets, respectively [14,15], and confers inhibition of negative transcriptional regulation of FGF-23. In contrast, inactivating mutations in FGF-23 or in the GALNT3 gene, the latter responsible for post-translational O-glycosylation of FGF-23, causes tumoural calcinosis, characterized by a reversed biochemical phenotype including hyperphosphataemia, elevated 1,25(OH)2D3 levels and extensive soft tissue and/or vascular calcification [16–18].

To further illuminate the complexity of FGF-23 actions, recent data suggest that FGF-23 directly inhibits the expression and secretion of parathyroid hormone (PTH). Our laboratory initially employed primary isolated bovine parathyroid cells to demonstrate direct FGF-23 signalling and suppressive effects on PTH mRNA levels and protein secretion in vitro [19], and these findings were elegantly substantiated in vivo by Ben-Dov and colleagues [20]. The role of FGF-23 in regulation of PTH in CKD is, at present, somewhat controversial and is subject to extensive investigation (see below).

To understand the biological actions of FGF-23, it is vital to elucidate the functional receptors mediating FGF-23 signalling. This has become a larger challenge than one at
first would have predicted. Despite intense efforts, many initial attempts to identify a specific FGF-23 receptor failed. A major breakthrough came from studies by Urakawa and co-workers, who demonstrated that type I membrane-bound alpha-Klotho (Klotho) directly interacts with FGF receptor 1c, converting it into a specific FGF-23 receptor [21]. In other words, Klotho functions as a permissive FGF-23 receptor co-factor and a determinant of FGF-23 tissue specificity. The importance of Klotho in FGF-23 signalling is supported by Klotho-null mice, which harbour nearly an identical biochemical phenotype compared to Fgf-23 knockout mice, despite exceptionally high circulating FGF-23 levels [9,11,21,22]. Thus, current data support that FGF-23, even at supraphysiological concentrations, likely has no impact on mineral metabolism without the presence of Klotho. The identification of the FGF receptors activated by FGF-23 in the kidney is critical for understanding the molecular mechanisms of FGF-23 actions on phosphate and vitamin D metabolism. Importantly, renal Klotho expression is largely confined to the distal tubules, which is also the site for initial FGF-23 binding and signalling [23,24]. However, renal phosphate reabsorption mainly occurs in the proximal tubules, and it is currently unknown how FGF-23 signalling in the distal tubule translates into decreased phosphate reabsorption in the proximal tubules.

FGF-23 action on organs other than kidney and parathyroid remain unanswered. As the predominant source of FGF-23 is osteocytes and osteoblasts [3,25], it was postulated that FGF-23 exerts local effects on bone. Available data are conflicting, but suggest that FGF-23 at least has an ‘intrinsic’ function in bone [26–28] (i.e. its expression changes cause downstream effects in the same cell leading to an altered bone phenotype), whereas direct endocrine effects are less likely given the complete absence of Klotho.

The principal physiological stimuli for increased FGF-23 expression both in vitro and in vivo are 1,25(OH)₂D₃ and high dietary phosphate intake [29–32]. Persistent hyperphosphataemia is also an effective trigger for FGF-23, although rapid changes in serum phosphate concentrations may not invoke an acute increment in serum FGF-23 levels [33]. It is therefore possible that FGF-23 responds to the net phosphate balance rather than the serum phosphate level, but experimental data supporting this hypothesis are scarce. Additionally, several studies indicate that PTH stimulates FGF-23 expression [34–36], closing the endocrine loop in the bone–parathyroid axis. In support, patients with primary hyperparathyroidism have mildly elevated FGF-23 levels in the face of hyperphosphataemia, but this could also be interpreted as an adaptive response to counteract the uncontrolled PTH secretion [37]. In hypoparathyroidism, a similar modest rise in FGF-23 is observed, which rather should be considered as a response to chronic hyperphosphataemia [38]. In this case, FGF-23 levels are not elevated to an extent that completely normalizes serum phosphate, presumably since this would have a deleterious impact on calcium metabolism through a further reduction in PTH as well as the 1,25(OH)₂D₃.

**FGF-23 and pathophysiology of CKD–MBD**

In CKD, the failing kidney is unable to adequately maintain mineral homeostasis, which initiates a series of events that inevitably lead to biochemical changes in serum, altered bone metabolism, vascular calcification and increased morbidity and mortality. All these characteristics are encompassed by the CKD–MBD. The triad of hyperphosphataemia, low 1,25(OH)₂D₃ and hypercalcemia are well-established triggers for increased PTH secretion and development of secondary hyperparathyroidism (SHPT). A more intricate question is what offsets this entire process in the earliest phase of CKD. Clearly, abnormalities in serum calcium and phosphate levels are not present until later CKD stages when estimated glomerular filtration rate (eGFR) falls below 25–30 ml/min. In fact, the maintenance of normal serum phosphate levels is presumably accomplished by a compensatory rise in FGF-23 [39,40], and this rise is detected already in the earliest stage of CKD, even in individuals without any clinically evident renal damage [41–43]. Increased FGF-23 levels in CKD must therefore primarily be considered as a result of a net positive phosphate balance, leading to increased renal phosphate excretion. Other potential explanations for the early rise in FGF-23 could be the release of unidentified FGF-23 stimulatory factors, or loss of a negative feedback factor(s) that normally suppress FGF-23, by the failing kidney. On the other hand, based on the physiological actions of FGF-23, this occurs at the expense of a reduction in 1,25(OH)₂D₃, explaining the observed calcitriol deficiency in early CKD. In other words, increased phosphaturia occurs at the expense of a reduction in 1,25(OH)₂D₃.

Before adequate experimental studies were performed, it was postulated that FGF-23 would stimulate, rather than decrease, PTH secretion. Indeed, a higher FGF-23 level in haemodialysis patients predicts future treatment-refractory SHPT [44]. Current data however suggest that the rise in FGF-23 is not merely a response to hyperphosphataemia but also to the increased PTH level [19,20]. Another puzzling observation is that transgenic mice over-expressing FGF-23 develop SHPT despite concomitant hypophosphataemia [7,8]. In this case, increased PTH levels are likely a rescue from severe hypocalcaemia due to extremely suppressed 1,25(OH)₂D₃. In support, deletion of the Pth gene in Hyp mice (a murine model of X-linked hypophosphataemic rickets suffering from increased FGF-23 expression) causes an embryonically lethal phenotype [45]. Several investigators have recently reported dramatically decreased expression levels of Klotho and FGF receptor 1c in hyperplastic parathyroid glands, leading to a parathyroid FGF-23 ‘resistance’ [46]. Since both FGF-23 and PTH increase in parallel with CKD development, it is possible that FGF-23 inhibits PTH secretion in early CKD, whereas this effect is attenuated or completely abolished in ‘FGF-23-resistant’ parathyroid glands. Future work will shed light on these important issues. A final remark is that it remains controversial whether FGF-23 or PTH rises first in the setting of progressive CKD. Some data from human cohort studies indicate that FGF-23 may rise before the devel-
Associations are shown for all subjects (n = 795) and >48 pg/mL, respectively. * adjusted for gender, serum phosphate, calcium, 25(OH)D3, PTH and eGFR. FGF-23 concentrations according to tertiles were <36 pg/mL, 36–93 pg/mL and >93 pg/mL, respectively. **P < 0.01; ***P < 0.001; reproduced with permission from [59].

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<th>Table 1. Circulating FGF-23 is associated with left ventricular hypertrophy (LVH) in a large, community-based cohort (PIVUS study)</th>
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<td><strong>Subjects with eGFR &lt;60 mL/min/1.73 m² (n = 164)</strong></td>
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<td>4.15** (1.74–9.92)</td>
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Adapted from [50] and [54]. Values shown are odds ratios (95% confidence interval). (A) Crude model; (B) adjusted for gender, systolic blood pressure, diastolic blood pressure, BMI, diabetes and hypertension; (C) adjusted for gender, serum phosphate, calcium, 25(OH)D3, PTH and eGFR. FGF-23 concentrations according to tertiles were <36 pg/mL, 36–48 pg/mL and >48 pg/mL, respectively. *P < 0.05; **P < 0.01; ***P < 0.001, reproduced with permission from [59].

opment of sHPT [42], but these observations need to be confirmed in additional studies. Speculatively, it is possible that a positive phosphate balance represents an early stimulator of FGF-23 in CKD, while an increased PTH level plays a later role in this process.

FGF-23, mortality and cardiovascular end points

Hyperphosphataemia, elevated PTH levels and 1,25(OH)₂D₃ deficiency are independently associated with increased mortality in CKD [47,48]. Since FGF-23 is a hormonal regulator of all these factors, it is not farfetched to believe that FGF-23 may represent a biomarker for a disordered mineral metabolism that is linked to survival. This important question was addressed by Gutierrez and co-workers, who explored the relation between FGF-23 and mortality in a prospective, nested case-control study of 400 incident haemodialysis patients [49]. Importantly, higher FGF-23 levels were strongly associated with increased risk of mortality both in crude- and multivariate-adjusted models, with the highest FGF-23 quartile reaching a nearly 6-fold higher risk than the lowest. FGF-23 was also superior to serum phosphate level in terms of predicting mortality, supporting the hypothesis that FGF-23 may represent a novel biomarker of a time-integrated phosphate burden, a more valuable parameter than a single serum phosphate measurement alone. The relation between FGF-23 and mortality has been validated in another smaller study [50], but it will nevertheless be valuable to confirm these findings in additional settings, especially in the general population given that a serum phosphate level, even within the normal range, is a predictor of mortality [51–53].

Since alterations in mineral metabolism are associated with increased cardiovascular risk, and are also a predominant cause of morbidity and mortality in CKD [47], another important question is whether FGF-23 is directly associated with cardiovascular risk factors. To test this hypothesis, we conducted a cross-sectional study employing a population-based cohort of ~1000 men and women aged 70 living in the community of Uppsala, Sweden. A majority of these patients were in CKD stage 2 with a mean eGFR of 73 ml/min/1.73 m², thus representing a valuable model of healthy individuals and early CKD. The rationale for this study was analogous to the Gutierrez report, i.e. to discern whether FGF-23 represents a biomarker of phosphate-induced cardiovascular toxicity in the community and in subjects with normal renal function. We found that a higher FGF-23 was linked to several dynamic measurements of vascular function, including arterial stiffness measured by pulse-wave velocity and endothelial dysfunction measured by an invasive forearm technique [43] in both crude- and multivariate-adjusted models. This supports that FGF-23 is linked to early changes in vascular function predisposing to an increased cardiovascular risk.

A subsample of the study population underwent a novel technique named whole-body magnetic resonance imaging angiography, which provides information about the degree of arterial stenosis as a surrogate marker of atherosclerosis in all major vascular territories. Analogous to the aortic calcification score or coronary artery calcium score, this method generates a ‘total-body’ atherosclerosis score. In agreement with previous findings, a higher FGF-23 level was associated with a higher atherosclerosis score, accentuating the role of FGF-23 as a marker of early cardiovascular changes [54]. It should be noted that FGF-23 in some studies has been linked to peripheral vascular calcification and/or coronary artery calcification score, whereas other reports have failed to show such an association [50,55–58].
This may, at least in part, be explained by imprecision and difficulties in standardizing the quantification of vascular calcification.

We further analysed the relation between FGF-23 and left ventricular hypertrophy, another large cardiovascular risk factor in CKD. This was also relevant because other members of the FGF family have been implicated in the pathogenesis of myocardial hypertrophy. Serum FGF-23 was positively associated with left ventricular mass index and increased risk of having left ventricular hypertrophy (Table 1). In particular, these associations were found in the highest FGF-23 tertile (>48 pg/mL) and were strengthened when restricted to subjects with eGFR <60 mL/min/1.73 m² [59]. Similar results were obtained by Gutierrez et al. [57] as well as in a Taiwanese haemodialysis study [60]. Interestingly, FGF-23 showed the strongest association with concentric left ventricular hypertrophy, which represents the most malignant form of left ventricular hypertrophy from a cardiovascular standpoint.

As a final comment to these studies, the associations between FGF-23, vascular dysfunction, atherosclerosis and left ventricular hypertrophy were all progressively strengthened in patients with a lower eGFR despite normal phosphate levels, supporting the hypothesis that FGF-23 may reveal information about phosphate-related toxicity that cannot be obtained by measurements of serum phosphate.

FGF-23: vascular protector, uraemic toxin or a plain biomarker?

Based on clinical and epidemiological outcome data, one could speculate that FGF-23 may not only serve as a biomarker of early cardiovascular changes due to altered mineral metabolism, but that FGF-23 itself directly targets the cardiovascular system (Fig. 1). Given that Klotho is neither expressed in the myocardium nor in blood vessels, it is unlikely that FGF-23 affects these tissues in an endocrine fashion. Another hypothesis is that the supraphysiological, or rather pharmacological, concentrations of FGF-23 present in many dialysis patients may induce unspecific, Klotho-independent, FGF receptor signalling. In this regard, FGF-receptor 1 is expressed in the heart [61,62], however, this hypothesis remains to be proven and certainly does not apply in the kidney, given the remarkably elevated levels of FGF-23 in Klotho-null mice that are unable to correct the hyperphosphataemia and high 1,25(OH)2D3 levels [21]. Similarly, patients with high circulating FGF-23 due to causes other than CKD, for example X-linked hypophosphataemic rickets and tumour-induced osteomalacia, do not suffer from a distinct increased cardiovascular risk.

Based on studies of FGF-23 as a potential mineralization inhibitor in bone, it is rather intriguing to speculate that FGF-23 could function as a local inhibitor of vascular calcification. Importantly, a preliminary report suggested that FGF-23 inhibits calcification in vascular smooth muscle cells in vitro; and these results were strengthened in an inflamed setting, which are often present in CKD patients [63]. Given the osteogenic transformation of vascular smooth muscle cells that occurs in atherosclerotic plaques, it is possible that FGF-23 may be locally expressed in the cardiovascular system. While a recent study failed to demonstrate FGF-23 expression in the heart or aorta of uraemic rats [64], it will nevertheless be important to determine if FGF-23 is expressed in the vasculature of CKD patients, especially at sites of extensive calcifications. It is currently unknown whether extraskeletal expression of FGF-23

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**Fig. 1.** FGF-23 functions as a hormonal regulator of mineral metabolism through direct effects renal phosphate reabsorption, 1,25-dihydroxyvitamin D₃ synthesis and PTH secretion. FGF-23 is associate with several cardiovascular risk factors, which hypothetically can be due to (i) indirect effects through hormonal control of mineral metabolism; (ii) local expression of FGF-23 in the vascular wall; (iii) direct, Klotho-independent effects of FGF-23 on the cardiovascular system.
contributes to the total amount of circulating FGF-23 in CKD. Finally, at least in early CKD, FGF-23 indirectly contributes to decreased vascular calcification through maintaining a normal serum phosphate level.

Conclusions

Although research surrounding FGF-23 metabolism initially was restricted to patients with rare hereditary disorders, it has progressively emerged as a key player in CKD–MBD. Multiple lines of evidence point to the importance of FGF-23 in normal and pathophysiological phosphate metabolism. Many questions remain elusive, yet FGF-23 holds promise at the very least as a biomarker of cardiovascular status and phosphate-related toxicity, both in CKD and in the general population. Future clinical trials targeting serum phosphate levels, even within in the normal range, will be of great importance to determine whether FGF-23 is a modifiable risk factor that can be translated into an earlier clinical management of disordered mineral metabolism in CKD.

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Conflict of interest statement. None declared.

References

5. Shimada T, Muto T, Urakawa I et al. Mutant FGF-23 responsible for autosomal dominant hypophosphatemic rickets is resistant to proteolytic cleavage and causes hypophosphatemia in vivo. Endocrinology 2002; 143: 3179–3182
26. A et al. 1alpha, 25-Dihydroxyvitamin D3 upregulates FGF23 gene expression in bone: the final link


57. Mirza MA, Larsson A, Melhus H et al. Serum intact FGF23 correlates significantly with left ventricular mass, hypertrophy and geometry in an elderly population. *Atherosclerosis* 2009 Dec; 207: 546–551


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