Published in final edited form as:

Curr Opin Nephrol Hypertens. 2016 July ; 25(4): 325–332. doi:10.1097/MNH.0000000000000232.

Inflammation regulates FGF23 production

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Abstract

Purpose of review—Fibroblast growth factor 23 (FGF23) is a hormone secreted by osteocytes and osteoblasts that regulates phosphorus and vitamin D homeostasis. FGF23 levels increase progressively in chronic kidney disease (CKD), and FGF23 excess might be a causal factor of left ventricular hypertrophy, CKD progression and death. Therefore, understanding the molecular mechanisms that control FGF23 production is a critical to design therapies to lower FGF23 levels. The present review focuses on the role of inflammatory stimuli on FGF23 regulation and summarizes recent studies that support a novel framework linking inflammation to FGF23 regulation.

Recent findings—Inflammation and iron deficiency, which are common occurrences in CKD have emerged as novel FGF23 regulators. Recent findings show that inflammation increases FGF23 production in bone through direct and iron-related indirect mechanisms. In these settings, Hypoxia Inducible Factor (HIF)-1 α orchestrates FGF23 transcription in response to inflammation and is primarily responsible for coordinating FGF23 production and cleavage.

Summary—We demonstrate that inflammation increases FGF23 production and may contribute to elevated FGF23 levels in CKD. Osseous HIF-1 α may represent a therapeutic target to lower FGF23 levels in CKD patients and minimize the negative consequences associated with FGF23 excess.

Keywords

FGF23; Hypoxia-inducible factor 1; inflammation; iron deficiency; chronic kidney disease

Introduction

FGF23 is a bone produced hormone that targets the kidney to regulate renal phosphate (Pi) handling and vitamin D metabolism. Primary FGF23 excess causes hypophosphatemia, aberrant vitamin D metabolism, impaired growth, and rickets/osteomalacia (1). Inversely, FGF23 deficiency results in hyperphosphatemia, excess 1,25(OH)₂D, and soft tissue calcifications (2-4). FGF23 production is regulated by systemic factors that affect mineral balance, mainly Vitamin D, PTH, phosphate and calcium (5-7). Since osteocytes and

osteoblasts are the main sources of circulating FGF23(8-11), bone alterations caused by local and systemic factors are susceptible to indirectly impact FGF23 production. Thus, FGF23 regulation might constitute a response to changes in bone and mineral metabolism to adjust vitamin D production, renal phosphate handling, and balance the mineral flux from bone.

Disordered bone and mineral metabolism is a common complication of CKD that begins early and worsens progressively as kidney function declines (12-14). CKD is the most common cause of elevated FGF23 and the clinical setting with the highest circulating levels (15). High FGF23 levels are strongly associated with greater risks of CKD progression, cardiovascular events, and mortality (16-21), and FGF23 excess is a causal factor for left ventricular hypertrophy (22, 23). These findings underscore the importance of designing therapies to lower FGF23 levels, but our current lack of understanding of the mechanisms regulating FGF23 production limits this approach. Recent findings suggest that inflammatory stimuli are major regulators of FGF23 production and could substantially contribute to the increase in FGF23 levels in CKD (24, 25), which we further discuss in this review.

FGF23 biology

FGF23 comprises a 24 amino acids hydrophobic signal sequence, an NH₂ terminal of 154 amino acids containing the FGF core homology region, and a characteristic 73 amino acids COOH-terminal domain.(26) In the bloodstream, the FGF23 protein circulates in distinct forms: a full-length mature form (25-FGF23-251) and cleaved shorter forms which mainly arise from proteolytic cleavage of full-length FGF23 at the 176RXXR179 site (27-29). O-glycosylation of FGF23 by UDP-N-acetyl- α -d-galactosamine:polypeptide N-acetylgalactosaminyl-transferase 3 (GALNT3) overlaps the 176RXXR179 cleavage site, and this posttranslational modification protects FGF23 from cleavage by furin-like proprotein convertases (26, 29). Indeed, GALNT3 or FGF23 mutations at glycosylation sites leads to low intact FGF23 levels with marked increase of processed COOH-terminal fragments in the circulation and results in hyperphosphatemic familial tumoral calcinosis (HFTC) (30, 31).

Additionally, phosphorylation of the serine 180 by family with sequence similarity 20, member C protein (FAM20C), inhibits O-glycosylation and promotes cleavage (32). In contrast with GALNT3 deletion, Fam20C knockout (KO) mice have impaired FGF23 cleavage leading to an increase in circulating bioactive FGF23 causing renal phosphate wasting and severe hypophosphatemic rickets(33). Thus, the levels of circulating and biologically active full-length FGF23 are regulated by GALNT3, FAM20C and furin-like proteases, respectively, responsible for the initiation of O-glycosylation, phosphorylation and proteolytic processing of FGF23.

FGF23 production and cleavage

It has become increasingly common to assess FGF23 production and cleavage non-invasively, in vivo, by using two different commercially available assays: the C-terminal FGF23 assay (cFGF23), which captures both intact FGF23 and its C-terminal fragments(34), and the intact FGF23 assay (iFGF23), which exclusively detects the intact hormone(27).

Using these 2 measures as a surrogate marker for FGF23 production and cleavage, we (35) and others (36) have shown that in multiple cases of pathological excess of FGF23 the elevation in circulating levels of intact FGF23 is caused by combined elevated FGF23 production and defective FGF23 cleavage.

The major example is found in patients suffering from autosomal dominant hypophosphatemic rickets (ADHR) caused by mutations at the cleavage site of FGF23 (28) that render FGF23 resistant to cleavage. In these patients, the manifestations of the disease can range from mild or no symptoms to late-onset of hypophosphatemic bone disease whereby increased FGF23 production is triggered by physiological states associated with iron deficiency(37), including puberty and pregnancy (38), and cleavage-resistant intact FGF23 accumulates in the bloodstream. As opposed to patients with ADHR, healthy subjects undergoing low iron states(39) display a proportional increase in FGF23 cleavage such that intact FGF23 levels remain unchanged or mildly affected. This strongly suggests that circulating FGF23 levels are tightly regulated by a balance between FGF23 transcription and cleavage under physiological conditions.

Inflammation and bone and mineral metabolism

Inflammation is part of the complex biologic response of any tissue to injury, infection, ischemia or autoimmune diseases. A variety of cytokines, including interleukin-1 β (IL-1 β), and acute phase proteins are released as a consequence of inflammation, in order to augment or attenuate the inflammatory response. Systemic inflammation alters bone and mineral metabolism at different levels. The mechanisms involved are complex and ultimately impact the bone remodeling cycle. Effects of acute and chronic inflammation on the skeleton are often very different. Acute inflammation generally increases both bone formation and bone resorption(40), while chronic inflammatory diseases of almost any cause are associated with bone loss, due to an uncoupling of bone formation from resorption in favour of excess resorption (41-45).

Multiple studies suggest that systemic inflammation may regulate FGF23 at least indirectly. Indeed, two of the main targets and regulators of FGF23, phosphorus and vitamin D, are strongly associated with systemic inflammation. Hypophosphatemia has long been reported to be associated with sepsis and correlates with sepsis severity(46, 47). Serum Pi is independently associated with inflammatory markers such as interleukin (IL)-6 (48), and dietary Pi loading dose dependently induces inflammation and increases serum tumor necrosis factor (TNF)- α levels in uremic rats(49). Vitamin D acts as an inhibitor of the inflammatory response (50) and decreases the mediators of systemic inflammation, such as interleukin-2 and TNF- α (51). Consequently, an inverse relation has been shown between vitamin D concentrations and C-reactive protein (CRP), a marker of inflammation, in both healthy subjects and patients with rheumatoid arthritis and frailty (52, 53). Finally, decreased vitamin D concentrations have been associated with an increased risk of developing autoimmune diseases, such as multiple sclerosis, rheumatoid arthritis, and type 1 diabetes (54, 55).

Inflammation regulates FGF23 production and cleavage

Associations between inflammatory markers and FGF23 have been reported in many inflammatory diseases (•56-62). Recently, we and others have shown that inflammatory cytokines are direct regulators of FGF23 production in cardiac fibroblasts (63) and osteoblasts (•24, •25). Using two separate models of acute inflammation resulting from the administration of either heat-killed *Brucella abortus* (BA) or IL-1 β in vivo, we observed approximately a ten-fold increase in *Fgf23* mRNA expression and serum cFGF23 while iFGF23 levels remained unchanged. The discrepancy between cFGF23 increase and the unaltered levels of intact hormone was due to concomitant increases in FGF23 production and cleavage. Indeed, co-administration of IL-1 β with a furin/furin-like protease inhibitor, which blocks FGF23 cleavage, significantly increased circulating iFGF23 compared to IL-1 β treatment alone. Also, furin inhibition did significantly increase intracellular and secreted iFGF23 in IL-1 β -treated cells versus controls only receiving IL-1 β . Collectively, these data suggest that inflammation is a powerful stimulator of FGF23 production and that FGF23 cleavage by furin/furin-like proteases plays a key role in maintaining adequate iFGF23 levels in response to a coinciding increase in circulating cFGF23.

Although the current data show that *Fgf23* expression and cFGF23 levels peak in the acute setting, *Fgf23* expression remains markedly elevated during chronic inflammation and, to different degrees, both cFGF23 and iFGF23 are increased during chronic inflammation. For unclear reasons, FGF23 processing is perturbed during chronic inflammation so that more biologically active FGF23 enters circulation. We speculate that prolonged states of amplified *Fgf23* expression overwhelm the cleavage capabilities within osteocytes but, at this stage, additional studies are needed to investigate the difference between chronic and acute inflammation on FGF23 production.

Direct and indirect effects of inflammation on FGF23 production

Systemic inflammation may affect FGF23 production indirectly. Indeed, inflammation-induced alterations of known regulators of FGF23, such as systemic calcium, phosphate and vitamin-D metabolism, might contribute to FGF23 elevation during inflammation. Paracrine stimuli secreted by osteoclasts, release of matrix bound components and/or local increase in phosphate and calcium due to inflammation-induced excessive bone resorption may also stimulate FGF23 (57, •64). In this context, excess calcium released by bone resorption may stimulate *Fgf23*(5) through calcineurin/NFAT pathway in osteoblasts(65). In support of this, inhibition of bone resorption reduces FGF23 levels in patients with osteogenesis imperfecta.⁽⁶⁶⁾

While these mechanisms are important, the major indirect effect of inflammation on FGF23 production is exerted through the regulation of iron metabolism. In vivo, the onset of inflammation is closely associated with the decline in circulating iron (•24, 67). As opposed to “true” iron deficiency, which reflects a state of low total body iron stores, “functional” iron deficiency is a consequence of chronic inflammation in which iron sequestration in the reticulo-endothelial system decreases the amount of circulating iron despite adequate total body iron stores (68, 69). In mice or humans, hypoferremia increases FGF23 transcription

but also cleavage, resulting in high circulating cFGF23 and normal or mildly elevated iFGF23 (•24, 36, 37, 39). In contrast, administration of iron to iron deficient patients decreases FGF23 (39). The effects of inflammation-induced functional iron deficiency on FGF23 are strikingly similar to those of true iron deficiency. Administration of exogenous murine hepcidin to WT mice, a peptide produced by the liver in response to inflammation, which increases iron sequestration and decreases gastrointestinal iron absorption, increased cFGF23 six hours post-injection (•24). However, these effects can only partially explain the FGF23 elevation in vivo and they do not account for the direct effects of inflammatory cytokines on FGF23 production in osteoblasts. Therefore, inflammation affects FGF23 production and cleavage directly through cytokine mediated mechanisms, and indirectly by limiting the amount of circulating iron.

Multiple mechanisms control FGF23 production during inflammation

One of the mechanisms at play during iron deficiency, is the normoxic stabilization of Hypoxia Inducible Factor (HIF)-1 α at the cellular level. Induction of HIF-1 α in osteoblasts/osteocytes, in response to iron-deficiency or hypoxia, increases FGF23 production (36, 70). In addition, HIF-1 α activation frequently occurs during inflammatory diseases (71) and many studies show that HIF-1 α functions in an adaptive manner by increasing ischemia tolerance and controlling excessive inflammation(72). In MC3T3-E1 osteoblast-like cells and bone marrow stromal cells, treatment with deferoxamine (DFO), an iron chelator, or IL-1 β significantly increased *Fgf23* mRNA expression (•24, 36). While both mechanisms involve HIF-1 α stabilization, DFO had no effect on *Hif-1 α* mRNA expression, but potently induced its nuclear abundance. Treatment with IL-1 β increased *Hif-1 α* mRNA expression and nuclear translocation in osteoblasts line suggesting that inflammation directly stimulates *Fgf23* mRNA expression through a HIF-1 α -dependent mechanism. In vivo, *Hif-1 α* mRNA expression was acutely elevated by IL-1 β but returned to baseline after four days of daily IL-1 β injections. While stabilization of HIF-1 α occurs when reduced cellular iron inhibit prolyl hydroxylase (73), mainly during iron deficiency and chronic inflammation, in acute inflammation there is an additional increase in *Hif-1 α* transcription, resulting in higher total HIF-1 α activity(74). In line with these findings, co-treatment of IL-1 β injected mice with a HIF-1 α inhibitor (2-methoxyestradiol (M2E2)), partially inhibited IL-1 β -induced increase in *Fgf23* promoter activity. Co-treatment with M2E2 also significantly reduced the abundance of nuclear HIF-1 α , cytoplasmic FGF23, and secreted FGF23 versus treatment with IL-1 β alone. In mice, M2E2 and an additional HIF-1 α inhibitor (BAY-87-2243) attenuated IL-1 β -induced increase in *Fgf23* mRNA expression, demonstrating a key role of HIF-1 α in FGF23 production.

Additional mechanisms, triggered by inflammation to increase FGF23 production are also involved. One study found that activation of nuclear factor kappa-light-chain-enhancer of activated B cells (NF- κ B), the prototypical proinflammatory pathway, potently increased FGF23 transcription in vitro (•25), and that NF- κ B inhibition also dramatically reduced FGF23 response to IL-1 β and TNF α . However, a 50 to 100-fold increase in FGF23 transcription remained unaccounted for even after NF- κ B inhibition. In line with our findings, this suggests the existence of several mechanisms controlling FGF23 production in response to inflammation, but these mechanisms might be closely interrelated. Indeed, NF-

κ B is a critical transcriptional activator of HIF-1 α (75). In turn hypoxia and HIF-1 α stabilization lead to NF- κ B activation (76) and amplify the NF- κ B pathway activation by increasing the expression and signaling of Toll-like receptors (TLR)s(77). Taken together these data suggest that normoxic HIF-1 α activation is a central mechanism to inflammation-dependent FGF23 production (Figure1).

Hif1a controls FGF23 production and cleavage

Beyond its role on FGF23 production, activation of HIF-1 α might also be responsible for the regulation of FGF23 cleavage. Indeed, HIF-1 α induces *furin* expression and increases the bioavailability of mature furin substrates in hypoxic conditions (78). In IL-1 β -injected mice, HIF-1 α inhibition results in increased circulating iFGF23. Conversely, increasing nuclear HIF-1 α abundance by prolyl-hydroxylase inhibitors increases *Fgf23* mRNA expression and circulating cFGF23. Finally, co-administration of furin inhibitors that partially inhibit FGF23 cleavage, with prolyl-hydroxylase inhibitors, that stabilize HIF-1 α , further increase iFGF23. Other findings show that pro-inflammatory stimuli acutely increased *Galnt3* mRNA which should protect FGF23 from cleavage in IDG-SW3 cells, while surprisingly detecting mainly FGF23 fragments (•25). These observations suggest that HIF-1 α coordinates cleavage during periods of FGF23 overproduction by increasing furin/furin-like proteases expression thus limiting the secretion of biologically active FGF23.

Inflammation: a potent stimulus for FGF23 in CKD

Chronic inflammation promotes renal, cardiac and vascular injury and is a major risk factor for systemic bone loss leading to fractures and substantial morbidity and mortality (79-81). Associations between kidney function and inflammation have been reported by multiple investigators (82-84), and inflammation has been shown to predict the long-term risk of developing chronic kidney disease (CKD) (85). FGF23 is associated with increased inflammatory burden in CKD (60) and we recently identified that FGF23-responsive genes are associated with renal damage and chronic inflammation (86). The Col4a3^{KO} mice, analog of human Alport's Syndrome and a validated model of progressive CKD, exhibit a time-dependent loss of kidney function in association with progressive elevation of cFGF23 and iFGF23 levels(14). Injection of IL-1 β in Col4a3^{KO} mice and WT mice resulted in a similar increase in cFGF23 and osseous *Fgf23* mRNA expression, and a similar decline in serum iron. Unexpectedly, serum iFGF23 was further increased in CKD mice when challenged acutely with IL-1 β , which suggests that FGF23 cleavage is impaired in CKD (•24). Given the prominent rise in iFGF23 in CKD compared to the very modest response in WT mice, it would seem that renal disease impairs FGF23 cleavage. Although it is unlikely that FGF23 cleavage occurs in the kidney (87), perhaps injured kidneys secrete a hormone that acts on bone to reduce intracellular FGF23 cleavage. Alternatively, a modification in *furin*, *fam20c* or *galnt3* expression levels in osteoblasts/osteocytes might constitute an initial adaptative mechanism to CKD settings. Regardless, these observations strongly suggest that systemic inflammation might be a potent stimulus for iFGF23 increase in CKD.

Conclusion

Inflammation and iron deficiency anemia are two hallmarks of CKD. Both inflammation and iron deficiency are potent inducers of FGF23 production and these pathways are of particular importance in a CKD setting. In a rather important twist, FGF23 also exhibits pro-inflammatory and immuno-modulatory effects, affecting macrophages and neutrophils (•88, ••89) suggesting the existence of a “feed-forward” loop in CKD (86), whereby the effects of inflammation and FGF23 amplify each other, leading to negative outcomes. Whether HIF-1 α , which regulates FGF23 production and cleavage, is also a target of excess FGF23, remains to be further investigated. Future exciting studies will determine if a dysfunctional osseous HIF-1 α could be a therapeutic target in CKD.

Acknowledgments

The authors would like to thank Dr. Aline Martin for critically reviewing the manuscript.

Financial support and sponsorship

This study was supported by NIH grant R01DK102815 to V. David.

Conflicts of interest

V. David receives research support from Keryx Biopharmaceuticals.

References and recommended reading

Publications of particular interest, published within the annual period of review, (18 months/ 2014- 2016) have been highlighted as:

- of special interest
- of outstanding interest

1. Martin A, David V, Quarles LD. Regulation and function of the FGF23/klotho endocrine pathways. *Physiological reviews*. 2012 Jan; 92(1):131–55. Epub 2012/02/03. eng. [PubMed: 22298654]
2. Shimada T, Kakitani M, Yamazaki Y, Hasegawa H, Takeuchi Y, Fujita T, et al. Targeted ablation of *Fgf23* demonstrates an essential physiological role of FGF23 in phosphate and vitamin D metabolism. *J Clin Invest*. 2004 Feb; 113(4):561–8. [PubMed: 14966565]
3. Benet-Pages A, Orlik P, Strom TM, Lorenz-Depiereux B. An FGF23 missense mutation causes familial tumoral calcinosis with hyperphosphatemia. *Hum Mol Genet*. 2005 Feb 1; 14(3):385–90. [PubMed: 15590700]
4. Stubbs JR, Liu S, Tang W, Zhou J, Wang Y, Yao X, et al. Role of hyperphosphatemia and 1,25-dihydroxyvitamin D in vascular calcification and mortality in fibroblastic growth factor 23 null mice. *J Am Soc Nephrol*. 2007 Jul; 18(7):2116–24. [PubMed: 17554146]
5. David D, Dai B, Martin A, Huang J, Han X, Quarles LD. Calcium regulates FGF-23 expression in bone. *Endocrinology*. 2013 Dec; 154(12):4469–82. [PubMed: 24140714]
6. Liu S, Tang W, Zhou J, Stubbs JR, Luo Q, Pi M, et al. Fibroblast growth factor 23 is a counter-regulatory phosphaturic hormone for vitamin D. *J Am Soc Nephrol*. 2006 May; 17(5):1305–15. [PubMed: 16597685]
7. Lopez I, Rodriguez-Ortiz ME, Almaden Y, Guerrero F, de Oca AM, Pineda C, et al. Direct and indirect effects of parathyroid hormone on circulating levels of fibroblast growth factor 23 in vivo. *Kidney Int*. 2011 Sep; 80(5):475–82. [PubMed: 21525854]
8. Liu S, Guo R, Simpson LG, Xiao ZS, Burnham CE, Quarles LD. Regulation of fibroblastic growth factor 23 expression but not degradation by PHEX. *J Biol Chem*. 2003 Sep 26; 278(39):37419–26. [PubMed: 12874285]

9. Liu S, Zhou J, Tang W, Jiang X, Rowe DW, Quarles LD. Pathogenic role of Fgf23 in Hyp mice. *Am J Physiol Endocrinol Metab*. 2006 Jul; 291(1):E38–49. [PubMed: 16449303]
10. Stubbs J, Liu S, Quarles LD. Role of fibroblast growth factor 23 in phosphate homeostasis and pathogenesis of disordered mineral metabolism in chronic kidney disease. *Semin Dial*. 2007 Jul-Aug;20(4):302–8. [PubMed: 17635819]
11. Yamashita T, Yoshioka M, Itoh N. Identification of a novel fibroblast growth factor, FGF-23, preferentially expressed in the ventrolateral thalamic nucleus of the brain. *Biochem Biophys Res Commun*. 2000 Oct 22; 277(2):494–8. [PubMed: 11032749]
12. Remuzzi G, Benigni A, Remuzzi A. Mechanisms of progression and regression of renal lesions of chronic nephropathies and diabetes. *J Clin Invest*. 2006 Feb.116:288–96. [PubMed: 16453013]
13. Isakova T, Wahl P, Vargas GS, Gutierrez OM, Scialla J, Xie H, et al. Fibroblast growth factor 23 is elevated before parathyroid hormone and phosphate in chronic kidney disease. *Kidney Int*. 2011 Jun; 79(12):1370–8. Epub 2011/03/11. eng. [PubMed: 21389978]
14. Stubbs JR, He N, Idiculla A, Gillihan R, Liu S, David V, et al. Longitudinal evaluation of FGF23 changes and mineral metabolism abnormalities in a mouse model of chronic kidney disease. *Journal of bone and mineral research : the official journal of the American Society for Bone and Mineral Research*. 2012 Jan; 27(1):38–46. Epub 2011/10/28. eng.
15. Wolf M. Update on fibroblast growth factor 23 in chronic kidney disease. *Kidney Int*. 2012 Oct; 82(7):737–47. Epub 2012/05/25. eng. [PubMed: 22622492]
16. Gutierrez OM, Mannstadt M, Isakova T, Rauh-Hain JA, Tamez H, Shah A, et al. Fibroblast growth factor 23 and mortality among patients undergoing hemodialysis. *N Engl J Med*. 2008 Aug 7; 359(6):584–92. [PubMed: 18687639]
17. Isakova T, Houston J, Santacruz L, Schiavenato E, Somarriba G, Harmon WG, et al. Associations between fibroblast growth factor 23 and cardiac characteristics in pediatric heart failure. *Pediatr Nephrol*. 2013 Oct; 28(10):2035–42. [PubMed: 23740037]
18. Kovesdy CP, Quarles LD. The role of fibroblast growth factor-23 in cardiorenal syndrome. *Nephron Clin Pract*. 2013; 123(3-4):194–201. [PubMed: 23942553]
19. Prie D, Forand A, Francoz C, Elie C, Cohen I, Courbebaisse M, et al. Plasma fibroblast growth factor 23 concentration is increased and predicts mortality in patients on the liver-transplant waiting list. *PLoS One*. 2013; 8(6):e66182. [PubMed: 23825530]
20. Touchberry CD, Green TM, Tchikrizov V, Mannix JE, Mao TF, Carney BW, et al. FGF23 is a novel regulator of intracellular calcium and cardiac contractility in addition to cardiac hypertrophy. *Am J Physiol Endocrinol Metab*. 2013 Apr 15; 304(8):E863–73. [PubMed: 23443925]
21. Wolf M, Molnar MZ, Amaral AP, Czira ME, Rudas A, Ujszaszi A, et al. Elevated fibroblast growth factor 23 is a risk factor for kidney transplant loss and mortality. *J Am Soc Nephrol*. 2011 May; 22(5):956–66. [PubMed: 21436289]
22. Faul C, Amaral AP, Oskoue B, Hu MC, Sloan A, Isakova T, et al. FGF23 induces left ventricular hypertrophy. *J Clin Invest*. 2011 Nov; 121(11):4393–408. [PubMed: 21985788]
- 23. Grabner A, Amaral AP, Schramm K, Singh S, Sloan A, Yanucil C, et al. Activation of Cardiac Fibroblast Growth Factor Receptor 4 Causes Left Ventricular Hypertrophy. *Cell Metab*. 2015 Dec 1; 22(6):1020–32. This study demonstrates that FGF23 promotes LVH by exclusively activating FGFR4 in cardiac myocytes. [PubMed: 26437603]
- 24. David V, Martin A, Isakova T, Spaulding C, Qi L, Ramirez V, et al. Inflammation and functional iron deficiency regulate fibroblast growth factor 23 production. *Kidney Int*. 2015 Nov 4. This study demonstrates that inflammation potently stimulates FGF23 production in-vitro and in-vivo, through a HIF-1 α dependent mechanism. FGF23 processing is also shown to be affected differently during acute inflammation, chronic inflammation, and chronic kidney disease.
- 25. Ito N, Wijanayaka AR, Prideaux M, Kogawa M, Ormsby RT, Evdokiou A, et al. Regulation of FGF23 expression in IDG-SW3 osteocytes and human bone by pro-inflammatory stimuli. *Mol Cell Endocrinol*. 2015 Jan 5.399:208–18. This study examines the role of various inflammatory stimuli and other local genes regulating FGF23 processing and production in osteocytes cultures. [PubMed: 25458698]

26. Kato K, Jeanneau C, Tarp MA, Benet-Pages A, Lorenz-Depiereux B, Bennett EP, et al. Polypeptide GalNAc-transferase T3 and familial tumoral calcinosis. Secretion of fibroblast growth factor 23 requires O-glycosylation. *J Biol Chem*. 2006 Jul 7; 281(27):8370–7.
27. Yamazaki Y, Okazaki R, Shibata M, Hasegawa Y, Satoh K, Tajima T, et al. Increased circulatory level of biologically active full-length FGF-23 in patients with hypophosphatemic rickets/osteomalacia. *J Clin Endocrinol Metab*. 2002 Nov; 87(11):4957–60. [PubMed: 12414858]
28. White KE, Carn G, Lorenz-Depiereux B, Benet-Pages A, Strom TM, Econs MJ. Autosomal-dominant hypophosphatemic rickets (ADHR) mutations stabilize FGF-23. *Kidney Int*. 2001 Dec; 60(6):2079–86. [PubMed: 11737582]
29. Shimada T, Muto T, Urakawa I, Yoneya T, Yamazaki Y, Okawa K, et al. Mutant FGF-23 responsible for autosomal dominant hypophosphatemic rickets is resistant to proteolytic cleavage and causes hypophosphatemia in vivo. *Endocrinology*. 2002 Aug; 143(8):3179–82. [PubMed: 12130585]
30. Bergwitz C, Banerjee S, Abu-Zahra H, Kaji H, Miyauchi A, Sugimoto T, et al. Defective O-glycosylation due to a novel homozygous S129P mutation is associated with lack of fibroblast growth factor 23 secretion and tumoral calcinosis. *J Clin Endocrinol Metab*. 2009 Nov; 94(11):4267–74. Epub 2009/10/20. eng. [PubMed: 19837926]
31. Yancovitch A, HersHKovitz D, Indelman M, Galloway P, Whiteford M, Sprecher E, et al. Novel mutations in GALNT3 causing hyperphosphatemic familial tumoral calcinosis. *Journal of bone and mineral metabolism*. 2011 Sep; 29(5):621–5. Epub 2011/02/25. eng. [PubMed: 21347749]
32. Tagliabracci VS, Engel JL, Wiley SE, Xiao J, Gonzalez DJ, Nidumanda Appaiah H, et al. Dynamic regulation of FGF23 by Fam20C phosphorylation, GalNAc-T3 glycosylation, and furin proteolysis. *Proceedings of the National Academy of Sciences of the United States of America*. 2014 Apr 15; 111(15):5520–5. Epub 2014/04/08. eng. [PubMed: 24706917]
33. Wang X, Wang S, Li C, Gao T, Liu Y, Rangiani A, et al. Inactivation of a novel FGF23 regulator, FAM20C, leads to hypophosphatemic rickets in mice. *PLoS genetics*. 2012; 8(5):e1002708. Epub 2012/05/23. eng. [PubMed: 22615579]
34. Jonsson KB, Zahradnik R, Larsson T, White KE, Sugimoto T, Imanishi Y, et al. Fibroblast growth factor 23 in oncogenic osteomalacia and X-linked hypophosphatemia. *N Engl J Med*. 2003 Apr 24; 348(17):1656–63. Epub 2003/04/25. eng. [PubMed: 12711740]
35. Martin A, David V, Li H, Dai B, Feng JQ, Quarles LD. Overexpression of the DMP1 C-terminal fragment stimulates FGF23 and exacerbates the hypophosphatemic rickets phenotype in Hyp mice. *Mol Endocrinol*. 2012 Nov; 26(11):1883–95. [PubMed: 22930691]
36. Farrow EG, Yu X, Summers LJ, Davis SI, Fleet JC, Allen MR, et al. Iron deficiency drives an autosomal dominant hypophosphatemic rickets (ADHR) phenotype in fibroblast growth factor-23 (Fgf23) knock-in mice. *Proceedings of the National Academy of Sciences of the United States of America*. 2011 Nov 15; 108(46):E1146–55. Epub 2011/10/19. eng. [PubMed: 22006328]
37. Imel EA, Peacock M, Gray AK, Padgett LR, Hui SL, Econs MJ. Iron modifies plasma FGF23 differently in autosomal dominant hypophosphatemic rickets and healthy humans. *J Clin Endocrinol Metab*. 2011 Nov; 96(11):3541–9. Epub 2011/09/02. eng. [PubMed: 21880793]
38. Breymann C, Honegger C, Holzgreve W, Surbek D. Diagnosis and treatment of iron-deficiency anaemia during pregnancy and postpartum. *Archives of gynecology and obstetrics*. 2010 Nov; 282(5):577–80. Epub 2010/06/26. eng. [PubMed: 20577752]
39. Wolf M, Koch TA, Bregman DB. Effects of iron deficiency anemia and its treatment on fibroblast growth factor 23 and phosphate homeostasis in women. *Journal of bone and mineral research : the official journal of the American Society for Bone and Mineral Research*. 2013 Aug; 28(8):1793–803. Epub 2013/03/19. eng.
40. Hardy R, Cooper MS. Bone loss in inflammatory disorders. *The Journal of endocrinology*. 2009 Jun; 201(3):309–20. Epub 2009/05/16. eng. [PubMed: 19443863]
41. Bischoff SC, Herrmann A, Goke M, Manns MP, von zur Muhlen A, Brabant G. Altered bone metabolism in inflammatory bowel disease. *The American journal of gastroenterology*. 1997 Jul; 92(7):1157–63. Epub 1997/07/01. eng. [PubMed: 9219790]
42. Compston JE, Vedi S, Croucher PI, Garrahan NJ, O'Sullivan MM. Bone turnover in non-steroid treated rheumatoid arthritis. *Ann Rheum Dis*. 1994 Mar; 53(3):163–6. [PubMed: 8154932]

43. Gough A, Sambrook P, Devlin J, Huissoon A, Njeh C, Robbins S, et al. Osteoclastic activation is the principal mechanism leading to secondary osteoporosis in rheumatoid arthritis. *J Rheumatol*. 1998 Jul; 25(7):1282–9. [PubMed: 9676757]
44. Robinson RJ, Iqbal SJ, Abrams K, Al-Azzawi F, Mayberry JF. Increased bone resorption in patients with Crohn's disease. *Aliment Pharmacol Ther*. 1998 Aug; 12(8):699–705. [PubMed: 9726381]
45. Uaratanawong S, Deesomchoke U, Lertmaharit S, Uaratanawong S. Bone mineral density in premenopausal women with systemic lupus erythematosus. *J Rheumatol*. 2003 Nov; 30(11):2365–8. [PubMed: 14677178]
46. Schwartz A, Brotfain E, Koyfman L, Kutz R, Gruenbaum SE, Klein M, et al. Association between Hypophosphatemia and Cardiac Arrhythmias in the Early Stage of Sepsis: Could Phosphorus Replacement Treatment Reduce the Incidence of Arrhythmias? *Electrolyte & blood pressure : E & BP*. 2014 Jun; 12(1):19–25. Epub 2014/07/26. eng. [PubMed: 25061469]
47. Schwartz A, Gurman G, Cohen G, Gilutz H, Brill S, Schily M, et al. Association between hypophosphatemia and cardiac arrhythmias in the early stages of sepsis. *European journal of internal medicine*. 2002 Oct. 13(7):434. Epub 2002/10/18. Eng. [PubMed: 12384132]
48. Navarro-Gonzalez JF, Mora-Fernandez C, Muros M, Herrera H, Garcia J. Mineral metabolism and inflammation in chronic kidney disease patients: a cross-sectional study. *Clinical journal of the American Society of Nephrology : CJASN*. 2009 Oct; 4(10):1646–54. Epub 2009/10/08. eng. [PubMed: 19808245]
49. Yamada S, Tokumoto M, Tatsumoto N, Taniguchi M, Noguchi H, Nakano T, et al. Phosphate overload directly induces systemic inflammation and malnutrition as well as vascular calcification in uremia. *American journal of physiology Renal physiology*. 2014 Jun 15; 306(12):F1418–28. Epub 2014/05/09. eng. [PubMed: 24808541]
50. Nagpal S, Na S, Rathnachalam R. Noncalcemic actions of vitamin D receptor ligands. *Endocrine reviews*. 2005 Aug; 26(5):662–87. Epub 2005/03/31. eng.
51. Lemire JM. Immunomodulatory role of 1,25-dihydroxyvitamin D3. *Journal of cellular biochemistry*. 1992 May; 49(1):26–31. Epub 1992/05/01. eng. [PubMed: 1644850]
52. Puts MT, Visser M, Twisk JW, Deeg DJ, Lips P. Endocrine and inflammatory markers as predictors of frailty. *Clinical endocrinology*. 2005 Oct; 63(4):403–11. Epub 2005/09/27. eng. [PubMed: 16181232]
53. Oelzner P, Muller A, Deschner F, Huller M, Abendroth K, Hein G, et al. Relationship between disease activity and serum levels of vitamin D metabolites and PTH in rheumatoid arthritis. *Calcified tissue international*. 1998 Mar; 62(3):193–8. Epub 1998/03/21. eng. [PubMed: 9501950]
54. Munger KL, Zhang SM, O'Reilly E, Hernan MA, Olek MJ, Willett WC, et al. Vitamin D intake and incidence of multiple sclerosis. *Neurology*. 2004 Jan 13; 62(1):60–5. Epub 2004/01/14. eng. [PubMed: 14718698]
55. Hillman L, Cassidy JT, Johnson L, Lee D, Allen SH. Vitamin D metabolism and bone mineralization in children with juvenile rheumatoid arthritis. *The Journal of pediatrics*. 1994 Jun; 124(6):910–6. Epub 1994/06/01. eng. [PubMed: 8201475]
- 56. Dounousi E, Torino C, Pizzini P, Cutrupi S, Panuccio V, D'Arrigo G, et al. Intact FGF23 and alpha-klotho during acute inflammation/sepsis in CKD patients. *European journal of clinical investigation*. 2016 Mar; 46(3):234–41. Epub 2016/01/06. Eng. This clinical study examines the regulation of FGF23 in CKD patients with acute sepsis and shows that increased proteolytic cleavage reduces circulating intact FGF23 at peak infection. [PubMed: 26728476]
57. Sato H, James Kazama J, Murasawa A, Otani H, Abe A, Ito S, et al. Serum Fibroblast Growth Factor 23 (FGF23) in Patients with Rheumatoid Arthritis. *Internal medicine (Tokyo, Japan)*. 2016; 55(2):121–6. Epub 2016/01/20. eng.
58. Hanks LJ, Casazza K, Judd SE, Jenny NS, Gutierrez OM. Associations of fibroblast growth factor-23 with markers of inflammation, insulin resistance and obesity in adults. *PLoS One*. 2015; 10(3):e0122885. Epub 2015/03/27. eng. [PubMed: 25811862]
- 59. Holecki M, Chudek J, Owczarek A, Olszanecka-Glinianowicz M, Bozentowicz-Wikarek M, Dulawa J, et al. Inflammation but not obesity or insulin resistance is associated with increased plasma fibroblast growth factor 23 concentration in the elderly. *Clinical endocrinology*. 2015 Jun;

82(6):900–9. Epub 2015/03/05. eng. This large cross-sectional study highlights the important relationship between inflammation and FGF23. Demonstrating an independent association between inflammation and FGF23 in elderly patients.

60. Munoz Mendoza J, Isakova T, Ricardo AC, Xie H, Navaneethan SD, Anderson AH, et al. Fibroblast growth factor 23 and Inflammation in CKD. *Clinical journal of the American Society of Nephrology : CJASN*. 2012 Jul; 7(7):1155–62. Epub 2012/05/05. eng. [PubMed: 22554719]
61. Iliopoulos D, Malizos KN, Oikonomou P, Tsezou A. Integrative microRNA and proteomic approaches identify novel osteoarthritis genes and their collaborative metabolic and inflammatory networks. *PLoS One*. 2008; 3(11):e3740. Epub 2008/11/18. eng. [PubMed: 19011694]
62. El-Hodhod MA, Hamdy AM, Abbas AA, Moftah SG, Ramadan AA. Fibroblast growth factor 23 contributes to diminished bone mineral density in childhood inflammatory bowel disease. *BMC gastroenterology*. 2012; 12:44. Epub 2012/05/04. eng. [PubMed: 22551310]
63. Yan L, Bowman MA. Chronic sustained inflammation links to left ventricular hypertrophy and aortic valve sclerosis: a new link between S100/RAGE and FGF23. *Inflammation and cell signaling*. 2014; 1(5) Epub 2014/01/01. Eng.
- 64. Yamazaki M, Kawai M, Miyagawa K, Ohata Y, Tachikawa K, Kinoshita S, et al. Interleukin-1-induced acute bone resorption facilitates the secretion of fibroblast growth factor 23 into the circulation. *Journal of bone and mineral metabolism*. 2015 May; 33(3):342–54. Epub 2014/07/06. eng. This study describes a relationship between inflammation, bone resorption, and FGF23 secretion. They report inflammation-induced bone resorption rather than increased production accounts for high circulating FGF23. [PubMed: 24996526]
65. Han X, Xiao Z, Quarles LD. Membrane and integrative nuclear fibroblastic growth factor receptor (FGFR) regulation of FGF-23. *J Biol Chem*. 2015 Apr 17; 290(16):10447–59. Epub 2015/03/11. eng. [PubMed: 25752607]
66. Kitaoka T, Namba N, Miura K, Kubota T, Ohata Y, Fujiwara M, et al. Decrease in serum FGF23 levels after intravenous infusion of pamidronate in patients with osteogenesis imperfecta. *Journal of bone and mineral metabolism*. 2011 Sep; 29(5):598–605. Epub 2011/02/24. eng. [PubMed: 21344299]
67. Braithwaite V, Prentice AM, Doherty C, Prentice A. FGF23 is correlated with iron status but not with inflammation and decreases after iron supplementation: a supplementation study. *International journal of pediatric endocrinology*. 2012; 2012(1):27. Epub 2012/10/27. eng. [PubMed: 23098062]
68. Zumbrennen-Bullough K, Babitt JL. The iron cycle in chronic kidney disease (CKD): from genetics and experimental models to CKD patients. *Nephrology, dialysis, transplantation : official publication of the European Dialysis and Transplant Association - European Renal Association*. 2014 Feb; 29(2):263–73. Epub 2013/11/16. eng.
69. Nemeth E, Ganz T. Anemia of inflammation. *Hematol Oncol Clin North Am*. 2014 Aug; 28(4): 671–81. vi. [PubMed: 25064707]
70. Clinkenbeard EL, Farrow EG, Summers LJ, Cass TA, Roberts JL, Bayt CA, et al. Neonatal iron deficiency causes abnormal phosphate metabolism by elevating FGF23 in normal and ADHR mice. *Journal of bone and mineral research : the official journal of the American Society for Bone and Mineral Research*. 2014 Feb; 29(2):361–9. Epub 2013/07/23. eng.
71. Colgan SP, Eltzschig HK. Adenosine and hypoxia-inducible factor signaling in intestinal injury and recovery. *Annual review of physiology*. 2012; 74:153–75. Epub 2011/09/29. eng.
72. Karhausen J, Furuta GT, Tomaszewski JE, Johnson RS, Colgan SP, Haase VH. Epithelial hypoxia-inducible factor-1 is protective in murine experimental colitis. *J Clin Invest*. 2004 Oct; 114(8): 1098–106. Epub 2004/10/19. eng. [PubMed: 15489957]
73. Knowles HJ, Mole DR, Ratcliffe PJ, Harris AL. Normoxic stabilization of hypoxia-inducible factor-1alpha by modulation of the labile iron pool in differentiating U937 macrophages: effect of natural resistance-associated macrophage protein 1. *Cancer research*. 2006 Mar 1; 66(5):2600–7. Epub 2006/03/03. eng. [PubMed: 16510578]
74. Blouin CC, Page EL, Soucy GM, Richard DE. Hypoxic gene activation by lipopolysaccharide in macrophages: implication of hypoxia-inducible factor 1alpha. *Blood*. 2004 Feb 1; 103(3):1124–30. Epub 2003/10/04. eng. [PubMed: 14525767]

75. Rius J, Guma M, Schachtrup C, Akassoglou K, Zinkernagel AS, Nizet V, et al. NF-kappaB links innate immunity to the hypoxic response through transcriptional regulation of HIF-1alpha. *Nature*. 2008 Jun 5; 453(7196):807–11. Epub 2008/04/25. eng. [PubMed: 18432192]
76. Cummins EP, Berra E, Comerford KM, Ginouves A, Fitzgerald KT, Seeballuck F, et al. Prolyl hydroxylase-1 negatively regulates IkappaB kinase-beta, giving insight into hypoxia-induced NFkappaB activity. *Proceedings of the National Academy of Sciences of the United States of America*. 2006 Nov 28; 103(48):18154–9. Epub 2006/11/23. eng. [PubMed: 17114296]
77. Kuhlicke J, Frick JS, Morote-Garcia JC, Rosenberger P, Eltzschig HK. Hypoxia inducible factor (HIF)-1 coordinates induction of Toll-like receptors TLR2 and TLR6 during hypoxia. *PLoS One*. 2007; 2(12):e1364. Epub 2007/12/27. eng. [PubMed: 18159247]
78. McMahon S, Grondin F, McDonald PP, Richard DE, Dubois CM. Hypoxia-enhanced expression of the proprotein convertase furin is mediated by hypoxia-inducible factor-1: impact on the bioactivation of proproteins. *J Biol Chem*. 2005 Feb 25; 280(8):6561–9. [PubMed: 15611046]
79. Stenvinkel P. Interactions between inflammation, oxidative stress, and endothelial dysfunction in end-stage renal disease. *Journal of renal nutrition : the official journal of the Council on Renal Nutrition of the National Kidney Foundation*. 2003 Apr; 13(2):144–8.
80. Stenvinkel P. Endothelial dysfunction and inflammation-is there a link? *Nephrology, dialysis, transplantation: official publication of the European Dialysis and Transplant Association - European Renal Association*. 2001 Oct; 16(10):1968–71.
81. Stenvinkel P. Inflammatory and atherosclerotic interactions in the depleted uremic patient. *Blood purification*. 2001; 19(1):53–61. [PubMed: 11114578]
82. Descamps-Latscha B, Herbelin A, Nguyen AT, Roux-Lombard P, Zingraff J, Moynot A, et al. Balance between IL-1 beta, TNF-alpha, and their specific inhibitors in chronic renal failure and maintenance dialysis. Relationships with activation markers of T cells, B cells, and monocytes. *Journal of immunology*. 1995 Jan 15; 154(2):882–92.
83. Herbelin A, Urena P, Nguyen AT, Zingraff J, Descamps-Latscha B. Elevated circulating levels of interleukin-6 in patients with chronic renal failure. *Kidney Int*. 1991 May; 39(5):954–60. [PubMed: 2067212]
84. Bolton CH, Downs LG, Victory JG, Dwight JF, Tomson CR, Mackness MI, et al. Endothelial dysfunction in chronic renal failure: roles of lipoprotein oxidation and pro-inflammatory cytokines. *Nephrology, dialysis, transplantation : official publication of the European Dialysis and Transplant Association - European Renal Association*. 2001 Jun; 16(6):1189–97.
85. Shankar A, Sun L, Klein BE, Lee KE, Muntner P, Nieto FJ, et al. Markers of inflammation predict the long-term risk of developing chronic kidney disease: a population-based cohort study. *Kidney Int*. 2011 Dec; 80(11):1231–8. Epub 2011/08/26. eng. [PubMed: 21866089]
86. Dai B, David V, Martin A, Huang J, Li H, Jiao Y, et al. A comparative transcriptome analysis identifying FGF23 regulated genes in the kidney of a mouse CKD model. *PLoS One*. 2012; 7(9):e44161. [PubMed: 22970174]
87. Christov M, Waikar SS, Pereira RC, Havasi A, Leaf DE, Goltzman D, et al. Plasma FGF23 levels increase rapidly after acute kidney injury. *Kidney Int*. 2013 Oct; 84(4):776–85. Epub 2013/05/10. eng. [PubMed: 23657144]
- 88. Han X, Li L, Yang J, King G, Xiao Z, Quarles LD. Counter-regulatory paracrine actions of FGF-23 and 1,25(OH)2 D in macrophages. *FEBS letters*. 2016 Jan; 590(1):53–67. Epub 2016/01/15. eng. This study examines effect of FGF23 and calcitriol on macrophage function. The pro-inflammatory actions of FGF23 and anti-inflammatory actions of calcitriol suggest FGF23 is an integral regulator of the immune response. [PubMed: 26762170]
- 89. Rossaint J, Oehmichen J, Van Aken H, Reuter S, Pavenstadt HJ, Meersch M, et al. FGF23 signaling impairs neutrophil recruitment and host defense during CKD. *J Clin Invest*. 2016 Mar 1; 126(3):962–74. Epub 2016/02/16. eng. This study shows that FGF23 inhibits chemokine-activated leukocyte arrest on the endothelium by signaling through FGFR2, and dampens host defense by direct interference with chemokine signaling and integrin activation. [PubMed: 26878171]

Key Points

- Herein we summarize recent studies that show compelling evidence of a close relationship between FGF23 production and processing and inflammation.
- We show that inflammation is a major stimulus for FGF23 production and metabolism and potentially a main component of FGF23 excess in chronic kidney disease.
- We explore different inflammation-driven mechanisms that stimulate FGF23 transcription and cleavage and highlight the importance of Hypoxia Inducible Factor 1 alpha as a central pathway controlling both FGF23 transcription and cleavage.

Figure 1. FGF23 regulation during inflammation

A. Acute Inflammation and consequent functional iron deficiency results in markedly increased HIF-1 α expression and stabilization HIF-1 α . Transcriptionally active HIF-1 α /HIF-1 β heterodimers translocate to the nucleus and bind hypoxia responsive elements (HRE) to simultaneously upregulate *Fgf23* and *Furin* expression. Inflammation also stimulates FGF23 production through alternate mechanisms. Furin overexpression counters the FGF23 elevation so that the majority of the secreted protein is cleaved. **B. Chronic inflammation** and functional iron deficiency induces HIF-1 α stabilization, but not HIF-1 α expression resulting in lower Furin/Furin-like protease activity compared to acute inflammation while the inflammatory stimulus driving *Fgf23* expression persists. Consequently, the cleavage is unable to match the rate of FGF23 production. In contrast to acute inflammation, more biologically active intact FGF23 enters circulation despite a lesser amount of total protein. **C. In CKD, chronic inflammation, true and functional iron deficiency** further stimulate FGF23 production. Additionally, unknown mechanisms reduce FGF23 cleavage in this setting. The net effect is a substantial elevation in circulating intact FGF23. We postulate that either Furin inhibition, increased O-glycosylation by Galnt3, or decreased phosphorylation by FAM20C reduce FGF23 cleavage in CKD.