Fibroblast Growth Factor 23 Regulation and Acute Kidney Injury

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Keywords

Fibroblast growth factor 23 · Acute kidney injury · Glycerol-3-phosphate · Lysophosphatidic acid

Abstract

Elevated fibroblast growth factor 23 (FGF23) levels are markers and potential mediators, of adverse outcomes in acute kidney injury (AKI). We recently identified glycerol-3-phosphate (G-3-P), a glycolysis byproduct, as a kidney-derived factor that circulates to bone and bone marrow and triggers FGF23 production in ischemic AKI. This kidney-to-bone signaling axis was further shown to require the conversion of G-3-P to lysophosphatidic acid (LPA) in bone marrow, followed by LPA signaling through the LPAR1 receptor. These findings highlight discrete steps potentially amenable to therapeutic targeting in conditions of FGF23 excess, although more work is required to determine the specificity and safety of targeting specific enzyme and receptor isoforms. Importantly, the initial metabolomic screen that identified a strong correlation between renal vein G-3-P and circulating FGF23 was conducted in human subjects undergoing elective catheterization, none with AKI. This raises the question of whether G-3-P might also modulate FGF23 homeostasis in patients with more mild or chronic decrements in kidney function, or under normal physiologic conditions – a question that is reinforced by a growing body of literature

highlighting functional roles for a range of circulating metabolites traditionally thought to function exclusively inside cells.

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Introduction

Fibroblast growth factor 23 (FGF23) levels rise rapidly with acute kidney injury (AKI) and are associated with the requirement for renal replacement therapy and death [1]. In addition to serving as an adverse prognostic marker, FGF23 may play a causal role through effects that extend beyond its traditional role in phosphate homeostasis, for example, by impacting the immune and cardiovascular systems [2, 3]. An improved understanding of why AKI stimulates FGF23 production outlines at least 2 potential opportunities: first, the development of treatment approaches to reduce FGF23 production in AKI and other pathophysiologic states of FGF23 excess and second,

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Fig. 1. A novel kidney-to-bone signaling axis modulates FGF23. AKI increases the production of glycerol-3-phosphate (G-3-P), which then circulates to bone and bm, where it is converted by GPAT to LPA. LPA action through an LPA receptor stimulates the production of intact, biologically active FGF23. FGF23, elevated fibroblast growth factor 23; AKI, acute kidney injury; G-3-P, glycerol-3-phosphate; LPA, lysophosphatidic acid; GPAT, glycerol-3-phosphate acyltransferase; bm, bone marrow.

Fig. 2. G-3-P is a byproduct of glycolysis. Glycolysis converts glucose to pyruvate through a series of intermediates that are shown. Pyruvate can then be used for oxidative metabolism within mitochondria (after conversion to acetyl-CoA) or can be reduced to lactate. G-3-P is produced from the reduction of the glycolytic intermediate dihydroxyacetone phosphate; it can also be produced by phosphorylation of glycerol. G-3-P, glycerol-3-phosphate.

the elucidation of regulatory mechanisms that may provide insight on FGF23 homeostasis under physiologic conditions.

Main Text

Minerals and related hormones including phosphate, calcium, 1,25(OH)₂D, and parathyroid hormone (PTH) all play a role in FGF23 homeostasis, but a comprehensive understanding of underlying mechanisms is lacking. For example, although exogenous phosphate is known to increase bone Fgf23 gene expression in vivo, phosphate does not appear to have a direct impact on FGF23 production in cultured bone cells. Recent studies raise the possibility that an important effect of phosphate is to reduce cleavage of intact FGF23 [4], or alternatively, that it is colloidal complexes of calcium phosphate and plasma protein fetuin-A that induce bone FGF23 production [5]. Further, recent years have yielded a proliferation of studies highlighting a role for nontraditional factors, including iron deficiency, erythropoietin, and inflammation in stimulating FGF23 production (as well as FGF23 cleavage) [6]. To what extent any of these factors – traditional or nontraditional – contribute to the rapid rise in FGF23 levels with AKI is uncertain.

Adding to this complexity, we recently identified glycerol-3-phosphate (G-3-P) as a kidney-derived metabolite that circulates to bone and bone marrow, where it is converted to lysophosphatidic acid (LPA), which then signals through the Lpar1 receptor to increase FGF23 production (shown in Fig. 1) [7]. To demonstrate potential clinical relevance, we measured plasma G-3-P levels in a matched case-control study of individuals who underwent cardiac surgery, where AKI was defined by a doubling of serum creatinine or need for renal replacement therapy. Whereas there was no difference in plasma G-3-P levels between cases and controls at baseline, immediately postoperative plasma G-3-P levels were significantly higher in AKI cases than controls (these samples were drawn in the operating room near the completion of surgery, before a rise in serum creatinine had occurred). Further, these postoperative plasma G-3-P levels had a stronger correlation with postoperative day 1 FGF23 levels than traditional FGF23 regulators, including phosphate, calcium, 1,25(OH)₂D, and PTH.

G-3-P is a byproduct of glycolysis (shown in Fig. 2), and thus is present throughout the body. We postulate that the increase in kidney G-3-P production in AKI is attributable to kidney ischemia, which is known to signifi-

cantly increase tubular epithelial cell glycolysis [8]. To explore signaling downstream of G-3-P, we used a standard model of kidney ischemia via renal artery clamping and reperfusion in mice. Using this approach, we documented elevations of glycolytic intermediates and G-3-P in kidney cortex and plasma, increases in G-3-P and LPA in bone marrow (no increase in LPA was observed in plasma), and a subsequent increase in FGF23 production. Further, we showed that the increase in FGF23 in this model of AKI is prevented if the conversion of G-3-P to LPA is inhibited or if the G-protein coupled receptor *Lpar1* is absent [7].

In a separate study, He et al. [9] have also shown that G-protein coupled receptor signaling in bone cells can stimulate FGF23 synthesis. More specifically, they found that ablation of the Gq/11 α -like, extra-large G α subunit (XL α S) causes FGF23 deficiency and hyperphosphatemia, with suppressed inositol 1,4,5-trisphosphate (IP3) and protein kinase C (PKC) levels in osteocytes. Across different biologic contexts, *Lpar1* has been shown to couple to Gi, Gq, and G12/13 α subunits, potentially placing *Lpar1* upstream of IP3/PKC signaling. However, whether LPA binding at *Lpar1* increases FGF23 production through this pathway is not known. Although further study is certainly warranted, *Lpar1* inhibition as a strategy to reduce FGF23 production has important limitations. First, we note that *Lpar1* deletion does not alter basal FGF23 levels in mice, only the response exogenous G-3-P or AKI, raising the possibility of redundant or compensatory signaling through other LPA receptors. And more importantly, the safety of targeting *Lpar1* may be limited by the wide-ranging expression and biologic function of the receptor [10].

Perhaps a more promising approach to reducing FGF23 production in AKI and other conditions of FGF23 excess is targeting the conversion of G-3-P to LPA. This reaction is catalyzed by the enzyme glycerol-3-phosphate acyltransferase (GPAT), which has 4 mammalian isoforms. In our study, we used a nonspecific inhibitor to block conversion of G-3-P to LPA in mice subjected to AKI. However, we also found that *Gpat2* is the main isoform expressed in bone and bone marrow, and that *Gpat2* knockdown in primary osteoblasts abrogated the stimulatory effect of G-3-P on FGF23 expression in vitro [7]. Corroborating these findings in vivo is a priority – unlike the other *Gpat* isoforms, *Gpat2* does not appear to play a major role in lipid metabolism, and thus may be a good candidate for therapeutic targeting, pending more definitive genetic or specific pharmacologic evidence that it catalyzes the conversion of G-3-P to LPA in bone and bone marrow [11].

Although both LPAR and GPAT inhibitors have been developed, molecules specific to LPAR1 and GPAT2 are not approved for clinical use. Of course, even if available, any attempt to lower FGF23 levels in patients would require careful consideration. An increase in FGF23 may reflect a compensatory response, for example, to mitigate hyperphosphatemia in progressive chronic kidney disease, such that reducing FGF23 levels is harmful [12]. Potential situations where the benefits of FGF23 reduction might outweigh risks could include situations where FGF23 is unable to promote urinary phosphate excretion, as with severe AKI or end-stage renal disease with minimal urine output, or genetic conditions of primary FGF23 excess [13].

In addition to identifying potential therapeutic targets in disease, our findings on FGF23 in AKI have the potential to shed new light on normal physiology. Importantly, the first clue that G-3-P may regulate FGF23 was from an analysis of kidney renal venous samples obtained from patients undergoing elective cardiac catheterization. More specifically, we performed metabolomic and proteomic profiling of renal venous plasma obtained from 17 individuals, and among the >300 metabolites and >1,300 proteins examined, the metabolite G-3-P had the strongest correlation with arterial FGF23. None of the individuals who underwent catheterization had AKI, and the mean estimated glomerular filtration rate (eGFR) across the sample was 66.6 mL/min per 1.73 m² [7]. This raises the question of whether mild and/or chronic kidney injury are also conditions where kidney-derived G-3-P modulates bone FGF23 production, or alternatively, whether other known FGF23 regulators act upstream of G-3-P.

Based on our metabolomics data, we know that ischemic AKI is one condition where increased glycolysis and G-3-P production are coupled. It is possible that chronic tissue ischemia could yield a similar result, but this requires experimental validation. Whether traditional or nontraditional FGF23 regulators can also impact G-3-P production is also uncertain, although the known stimulatory impact of hypoxia-inducible factors (HIFs) on glycolysis is noteworthy, given the interactions between iron deficiency, erythropoietin, inflammation, and HIF action [6]. Intriguingly, SGLT2 inhibitors have been found to increase both FGF23 and phosphate levels, an unanticipated finding [14]. The impact of SGLT2 inhibition on proximal tubule metabolism is the subject of ongoing investigation in the field, but an impact on kidney glycolysis is plausible given its fundamental impact on apical glucose uptake. Importantly, SGLT2 expression is restricted

to the kidney. By contrast, glycolysis and G-3-P production are ubiquitous processes. Thus, any mechanism that would purport to modulate FGF23 via kidney G-3-P production would warrant a similar organ-specific explanation.

If G-3-P does in fact play a role in FGF23 homeostasis under physiologic conditions, it would add to the growing list of circulating metabolites that have been ascribed hormone-like functions [15]. Numerous molecules traditionally thought to function exclusively within cells, including citric acid cycle intermediates, short chain fatty acids, bile acids, aromatic amino acids, and lactate have been found to play systemic roles in energy metabolism, immune tolerance, and even blood pressure. In many cases, these molecules have been found to be specific ligands for previously “orphan” G-protein coupled receptors or nuclear receptors. As with the G-3-P, LPA, and LPAR1 signaling axis, more work will be required to explore the therapeutic and physiological implications of these findings.

Conclusion

Kidney-derived G-3-P is a novel factor that can trigger FGF23 production in bone and bone marrow, with demonstration of potential clinical relevance in AKI. Downstream of kidney release, the conversion of G-3-P to LPA and LPA signaling through LPAR1 represent 2 nodes where this signaling axis may be interrupted. G-3-P is a byproduct of glycolysis, and its production is significantly increased when glycolysis is stimulated, as with ischemic AKI. Future studies should seek to determine whether other factors, including traditional and nontraditional FGF23 regulators, may lie upstream of kidney glycolysis and G-3-P production under physiological conditions.

Conflict of Interest Statement

The authors have no conflicts of interest to declare.

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