

Characterizing Splenectomy-Induced Exaggerated Bone Loss in a Pre-Clinical beta-Thalassemia Mouse Model: Role of Elevated FGF23

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INTRODUCTION: Patients with hemolytic disorders like β -thalassemias often experience osteoporosis and fragility fractures. Surgical splenectomy, aimed at maintaining hemoglobin levels, has been linked to increased osteoporosis risk in thalassemic patients. However, the underlying mechanism(s) remain unclear, and we are still lacking an appropriate animal model represents the post-splenectomy bone loss. This study investigates the impact of splenectomy on bone homeostasis in a thalassemic mouse model and explores potential mechanisms.

METHODS: A heterozygous mouse model of β^0 -thalassemia ($Hbb^{th3/+}$) was utilized. Surgical splenectomy was performed at 4 weeks on thalassemic and WT mice. Bone density, geometry, and bone histomorphometry were assessed at 10 weeks post-splenectomy via micro-CT, static and dynamic bone morphometric analyses. Circulating intact FGF23 (iFGF23) was measured via ELISA. The expression of FGF23 in bone were evaluated by immune-histochemistry staining of FGF23 on whole bones. Furthermore, whole bones were separated to bone matrix and bone marrow. The expressions of FGF23 from bone matrix and bone marrow were assessed by RT-qPCR and Western blot separately. To investigate the causative role of high circulating FGF23 on bone loss induced by splenectomy, the function of circulating FGF23 was blocked by administering FGF23 neutralization antibody (FGF23Ab, 10mg/kg, Amgen Inc). bone morphometric changes were evaluated by MciroCT morphometric analysis.

RESULTS: Th3+/- mice displayed progressive bone loss and morphological changes. Splenectomy exacerbated these deteriorative changes in thalassemic mice, characterized by decreased bone volume fraction, trabecular number, and trabecular thickness in vertebrae and proximal tibia; decreased bone area and increased marrow area in cortical bone of mid-shaft of femur (**Fig. 1A**). Histomorphometric analyses revealed reduced osteoblast and increased osteoclast surfaced in thalassemic mice, which were further affected by splenectomy (**Fig. 1B**). Elevated circulating intact FGF23 (iFGF23) levels were observed in thalassemic mice, and further elevated post-splenectomy (**Fig. 1C**). FGF23 expression in bone matrix and bone marrow was higher in thalassemic mice, with a significant increase from the bone marrow after splenectomy (**Fig. 1D**). Furthermore, FGF23 neutralization antibody treatment reversed splenectomy-induced bone loss (**Fig. 1E**).

DISCUSSION: This study unveils a novel pathological role of elevated FGF23 in contributing to bone abnormalities in β -thalassemia, particularly after splenectomy. The findings underscore the importance of understanding the interplay between hemolytic disorders, bone homeostasis, and surgical interventions, shedding light on potential therapeutic strategies for bone health in thalassemia patients.

SIGNIFICANCE/CLINICAL RELEVANCE: This study shed light on the etiology of poor bone health in thalassemia and revealed bone marrow derived FGF23 as a therapeutic target to prevent splenectomy induced bone loss in thalassemia.

IMAGES AND TABLES:

