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Pathophysiology

"BONE RESORPTION MECHANISMS IN THALASSEMIA: A FOCUS ON OSTEOCLASTS, CTX LEVELS, AND VEGF's ROLE IN PATHOGENESIS"

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ABSTRACT Thalassemia, a genetic hemoglobin disorder, is associated with numerous complications, including bone disorders such as osteoporosis. Osteoclast-mediated bone resorption plays a key role in the pathophysiology of thalassemia-associated bone loss. Biomarkers such as C-terminal telopeptide (CTX) are elevated in thalassemia, reflecting increased bone turnover. Vascular endothelial growth factor (VEGF), known for its role in angiogenesis, also contributes to the regulation of bone remodeling and is implicated in the pathogenesis of bone disorders in thalassemia. This review aims to explore the mechanisms of bone resorption in thalassemia, focusing on osteoclast biology, the significance of CTX levels, and VEGF's role in exacerbating bone loss.

KEYWORDS : *Bone mineral density (BMD), Vascular endothelial growth factor (VEGF), Osteoclast, osteoporosis, C-terminal telopeptide (CTX)*

INTRODUCTION

Thalassemia is a hereditary disorder characterized by defective hemoglobin production due to mutations in the genes encoding the alpha or beta globin chains. The resulting ineffective erythropoiesis leads to chronic anemia, which induces a cascade of compensatory mechanisms that impact multiple organ systems, including the skeletal system. One of the most prominent complications in patients with thalassemia is osteoporosis, characterized by reduced bone density and increased fracture risk, particularly in patients with thalassemia major and intermediate.

While the pathogenesis of bone disease in thalassemia is multifactorial, bone resorption, mediated by osteoclasts, is a critical contributor. In this context, this review will explore the underlying mechanisms driving bone resorption in thalassemia, with a particular focus on osteoclast activity, the relevance of CTX as a biomarker of bone turnover, and the emerging role of VEGF in modulating bone resorption. By elucidating these factors, we aim to offer a comprehensive understanding of the pathophysiology of bone loss in thalassemia, which may inform future therapeutic strategies.

Pathophysiology Of Bone Disease In Thalassemia

Overview of Bone Remodeling

Bone homeostasis is a dynamic process involving the coordinated activity of osteoclasts (responsible for bone resorption) and osteoblasts (responsible for bone formation). These cells work in concert to maintain bone strength and density through a process known as bone remodeling, which involves resorption of old or damaged bone and the formation of new bone.

In healthy individuals, the balance between bone resorption and formation ensures skeletal integrity. However, in pathological conditions such as thalassemia, this balance is disrupted, resulting in excessive bone resorption that surpasses bone formation, leading to osteoporosis. This imbalance is particularly evident in patients with thalassemia major, who often exhibit increased osteoclast activity and decreased osteoblast function, contributing to accelerated bone loss.

Thalassemia-Induced Bone Disease: Clinical Manifestations

Osteoporosis in thalassemia presents with a variety of clinical manifestations, including reduced bone mineral density (BMD), deformities such as kyphosis and scoliosis, and an increased risk of fractures, particularly in weight-bearing bones like the spine, hips, and long bones. These skeletal complications often begin in childhood and progress with age, leading to significant morbidity in affected individuals. Early diagnosis and treatment of bone disease in thalassemia are critical for preventing long-term skeletal complications.

Osteoclast Biology and Function in Thalassemia

Osteoclast Differentiation and Activation

Osteoclasts are multinucleated cells derived from the hematopoietic lineage of monocytes and macrophages. The differentiation of osteoclast precursors into mature, bone-resorbing cells is regulated by several factors, including receptor activator of nuclear factor kappa-B ligand (RANKL), macrophage colony-stimulating factor (M-CSF), and osteoprotegerin (OPG).

In the bone microenvironment, RANKL binds to its receptor, RANK, on osteoclast precursors, promoting their differentiation into mature osteoclasts. M-CSF enhances the proliferation and survival of osteoclast precursors, while OPG, a decoy receptor for RANKL, inhibits osteoclastogenesis by preventing RANKL from binding to RANK.

Enhanced Osteoclastogenesis In Thalassemia

In thalassemia, the pathogenesis of bone disease is driven by enhanced osteoclastogenesis, leading to excessive bone resorption. Several mechanisms contribute to increased osteoclast activity in thalassemia:

1.Hypoxia: Chronic anemia in thalassemia results in tissue hypoxia, which induces the expression of hypoxia-inducible factors (HIFs) that promote osteoclast differentiation. Hypoxia also stimulates the production of RANKL by osteoblasts and stromal cells, further enhancing osteoclastogenesis [1, 2].

2. Iron Overload: Repeated blood transfusions in thalassemia patients result in iron overload, which contributes to oxidative stress and increased production of pro-inflammatory cytokines such as tumor necrosis factor-alpha (TNF-α) and interleukin-6 (IL-6). These cytokines are potent inducers of osteoclast formation and activity [3, 4].

3. Increased Inflammatory Cytokines: Chronic inflammation in thalassemia is characterized by elevated levels of pro-inflammatory cytokines, including TNF-α, IL-1, and IL-6, which promote osteoclast differentiation and activity through the RANKL pathway [5, 6].

Osteoclast-Mediated Bone Resorption

Once differentiated, osteoclasts attach to the bone surface and create a sealed resorption zone, where they secrete hydrogen ions and proteolytic enzymes such as cathepsin K to degrade the mineralized bone matrix. This process results in the release of calcium and phosphate into the bloodstream and the breakdown of collagen, which is released as degradation products such as C-terminal telopeptide (CTX) [7, 8].

In thalassemia, the excessive activity of osteoclasts leads to increased bone resorption, resulting in reduced bone mass and structural deterioration of the skeleton.

C-Terminal Telopeptide (CTX) as a Biomarker of Bone Turnover

Role Of CTX In Bone Turnover

C-terminal telopeptide (CTX) is a collagen degradation product that is released into the bloodstream during bone resorption. It is considered a reliable marker of bone turnover, as elevated CTX levels indicate increased osteoclast activity and heightened bone resorption. Serum or urinary CTX measurements are commonly used to assess bone resorption in various metabolic bone disorders, including osteoporosis [9, 10].

Elevated CTX Levels in Thalassemia

Numerous studies have demonstrated that CTX levels are significantly elevated in patients with thalassemia, reflecting increased osteoclast-mediated bone resorption. Elevated CTX levels have been shown to correlate with reduced bone mineral density (BMD) and increased fracture risk in these patients [11, 12].

In thalassemia, several factors contribute to elevated CTX levels:

1. Osteoclast Activation: As previously discussed, enhanced osteoclastogenesis in thalassemia results in increased bone resorption, leading to higher levels of CTX in the bloodstream.

2. Iron Overload and Oxidative Stress: Iron overload in thalassemia generates oxidative stress, which promotes osteoclast activity and increases CTX release [13].

3.Inflammation: Chronic inflammation in thalassemia, driven by elevated pro-inflammatory cytokines, further enhances osteoclast activity and CTX production [14].

Clinical Significance of CTX in Thalassemia

Monitoring CTX levels in thalassemia patients provides valuable insights into the degree of bone turnover and the severity of bone loss. Elevated CTX levels may serve as a biomarker for assessing the progression of osteoporosis in thalassemia and the effectiveness of therapeutic interventions aimed at reducing bone resorption.

In clinical practice, CTX measurements are often used to evaluate the response to anti-resorptive therapies such as bisphosphonates, which aim to reduce osteoclast activity and bone resorption. Studies have shown that bisphosphonate treatment in thalassemia patients can significantly reduce CTX levels, indicating decreased bone resorption and improved bone health [15, 16].

Vascular Endothelial Growth Factor (VEGF) and Its Role in Bone Resorption

VEGF in Bone Physiology

Vascular endothelial growth factor (VEGF) is a key regulator of angiogenesis, the process of new blood vessel formation. In addition to its role in angiogenesis, VEGF plays a critical role in bone remodeling by influencing both osteoclast and osteoblast function. VEGF promotes the recruitment and differentiation of osteoclast precursors, thereby enhancing bone resorption [17, 18].

Hypoxia and VEGF Upregulation in Thalassemia

Chronic anemia in thalassemia results in tissue hypoxia, which stimulates the production of VEGF. Hypoxia-inducible factor-1 (HIF-1) is a transcription factor that is upregulated in response to low oxygen levels and promotes the expression of VEGF. Elevated VEGF levels in thalassemia contribute to increased angiogenesis and bone resorption by enhancing osteoclastogenesis [19, 20].

VEGF-Mediated Osteoclast Activity

VEGF exerts its effects on osteoclasts by interacting with the RANKL/RANK pathway, which is essential for osteoclast differentiation and activation. VEGF enhances the production of RANKL by osteoblasts and stromal cells, promoting osteoclastogenesis and bone resorption. In thalassemia, the increased expression of VEGF in response to hypoxia and inflammation contributes to the excessive activation of osteoclasts and the resulting bone loss [21, 22].

VEGF As A Therapeutic Target In Thalassemia-associated Bone Loss

Given the role of VEGF in promoting bone resorption, targeting VEGF signaling represents a potential therapeutic approach for reducing bone loss in thalassemia. Anti-VEGF therapies, such as bevacizumab and aflibercept, have been developed to inhibit VEGF activity and are currently used in the treatment of various cancers and ocular diseases. These agents may hold promise in reducing osteoclast activity and preventing bone loss in patients with thalassemia [23, 24].

Additional Factors Contributing to Bone Resorption in Thalassemia

Iron Overload And Oxidative Stress

Iron overload is a major complication in thalassemia, resulting from repeated blood transfusions and increased intestinal iron absorption. Excessive iron deposition in various tissues, including bone, leads to the generation of reactive oxygen species (ROS) and oxidative stress. Oxidative stress plays a critical role in promoting osteoclast activity and bone resorption, as ROS enhance osteoclast differentiation and activity through the activation of signaling pathways such as nuclear factor-kappa B (NF-κB) and mitogen-activated protein kinases (MAPKs) [25, 26].

In addition to promoting osteoclast activity, iron overload impairs osteoblast function, further tipping the balance toward bone resorption. Studies have shown that iron inhibits osteoblast proliferation and differentiation, leading to reduced bone formation and contributing to the overall bone loss observed in thalassemia [27, 28].

Endocrine Dysfunction

Endocrine dysfunction is common in thalassemia, particularly in patients with thalassemia major who have undergone repeated blood transfusions and are at risk of developing iron overload. Hypogonadism, characterized by reduced sex hormone levels, is one of the most prevalent endocrine complications in thalassemia and is strongly associated with bone loss. Estrogen and testosterone play critical roles in regulating bone metabolism by inhibiting osteoclast activity and promoting osteoblast function [29].

In thalassemia, untreated hypogonadism leads to increased osteoclast activity and bone resorption, contributing to the development of osteoporosis. Hormone replacement therapy (HRT) has been shown to reduce bone resorption and improve bone density in hypogonadal patients with thalassemia [30, 31].

Chronic Inflammation

Chronic inflammation is a hallmark of thalassemia, driven by ongoing hemolysis, iron overload, and increased erythropoietic activity. Elevated levels of pro-inflammatory cytokines, including TNF-α, IL-1, and IL-6, promote osteoclast differentiation and activity by upregulating RANKL expression and activating osteoclast precursors [32, 33].

In addition to promoting osteoclastogenesis, chronic inflammation in thalassemia inhibits osteoblast function, leading to reduced bone formation and further exacerbating bone loss. Anti-inflammatory therapies, such as TNF-α inhibitors, have shown potential in reducing osteoclast activity and improving bone health in inflammatory conditions and may represent a therapeutic option for thalassemia-associated bone disease [34, 35].

Therapeutic Approaches for Bone Resorption in Thalassemia

Bisphosphonates

Bisphosphonates are the most widely used anti-resorptive agents for the treatment of osteoporosis in thalassemia. These drugs inhibit osteoclast activity by inducing apoptosis in osteoclasts, thereby reducing bone resorption. Several clinical studies have demonstrated the efficacy of bisphosphonates in increasing bone mineral density (BMD) and reducing fracture risk in patients with thalassemia [36, 37]. Bisphosphonate therapy has also been shown to reduce CTX levels, indicating decreased bone turnover and improved bone health. Long-term bisphosphonate treatment has been associated with sustained improvements in BMD and reductions in fracture incidence in thalassemia patients [38, 39].

Denosumab

Denosumab is a monoclonal antibody that targets RANKL, inhibiting its interaction with RANK on osteoclast precursors and thereby reducing osteoclast differentiation and activity. Denosumab has been shown to effectively reduce bone resorption and increase bone density in patients with osteoporosis and other metabolic bone disorders [40, 41].

Given the role of RANKL in promoting osteoclastogenesis in thalassemia, denosumab represents a promising therapeutic option for reducing bone resorption in this population. However, further studies are needed to evaluate the long-term efficacy and safety of denosumab in patients with thalassemia [42, 43].

Iron Chelation Therapy

Iron chelation therapy is a cornerstone of treatment in thalassemia, aimed at reducing iron overload and preventing its complications, including bone loss. By reducing iron-induced oxidative stress and inflammation, iron chelation therapy can mitigate osteoclast activity and bone resorption [44, 45].

Studies have shown that iron chelation therapy with agents such as deferasirox and deferoxamine improves bone mineral density and reduces bone turnover markers in thalassemia patients. Early initiation of iron chelation therapy is critical for preventing iron-related skeletal complications [46, 47].

Hormone Replacement Therapy (HRT)

In patients with thalassemia who develop hypogonadism, hormone replacement therapy (HRT) is essential for maintaining bone health. Estrogen and testosterone replacement therapies have been shown to reduce bone resorption, increase bone density, and prevent fractures in hypogonadal individuals with thalassemia [48, 49].

HRT should be initiated early in the course of hypogonadism to prevent the development of osteoporosis and other skeletal complications. In addition to improving bone health, HRT may also have positive effects on overall quality of life in thalassemia patients [50, 51].

Anti-VEGF Therapies

Given the role of VEGF in promoting osteoclastogenesis and bone resorption, anti-VEGF therapies represent a novel therapeutic approach for treating bone loss in thalassemia. By inhibiting VEGF signaling, these agents may reduce osteoclast activity and prevent the progression of osteoporosis [52, 53].

Anti-VEGF therapies, such as bevacizumab and aflibercept, are currently used in the treatment of various cancers and ocular diseases. Preliminary studies suggest that these agents may also have potential in reducing bone resorption in conditions characterized by excessive osteoclast activity, such as thalassemia [54, 55].

Polyphenols/Flavonoids

Thalassemia patients commonly experience heightened oxidative stress and iron overload, which contribute to bone resorption and impaired bone mineral density via reactive oxygen species disrupting bone marrow microenvironments and accelerating osteoclast activity. Incorporating polyphenol-rich antioxidants such as resveratrol and curcumin into an antioxidant-rich dietary strategy offers a promising adjuvant approach. Resveratrol, a stilbene found in grapes and berries, has been shown to scavenge ROS, enhance osteogenesis, suppress osteoclast formation, and increase bone mineral density by activating SIRT1/Runx2 and modulating NF-κB signaling in resorptive bone disease models.[56-57]

Curcumin, a phenolic compound derived from turmeric, exhibits iron-chelating, anti-osteoclastogenic, and anti-inflammatory properties and protects osteoblasts from apoptosis, thus helping limit bone loss .[58] Additionally, in β-thalassemia/HbE patients, curcuminoid supplementation (e.g., 500 mg/day) over 12 months improved oxidative stress markers, elevated antioxidant enzyme levels, and reduced non-transferrin-bound iron. These systemic benefits may indirectly support healthier bone metabolism.[59]

Together, this dietary antioxidant approach centered on resveratrol and curcumin may help counteract thalassemia-associated bone resorption through attenuating oxidative damage, iron toxicity, and osteoclast activity while supporting osteoblast function and bone remodeling.

CONCLUSION

Bone resorption in thalassemia is a complex process driven by multiple factors, including enhanced osteoclast activity, elevated CTX levels, and dysregulated VEGF signaling. These mechanisms are further exacerbated by iron overload, chronic inflammation, and endocrine dysfunction, all of which contribute to accelerated bone loss and the development of osteoporosis.

Current treatment strategies for bone disease in thalassemia focus on reducing osteoclast activity and addressing the underlying causes of bone loss, such as iron overload and hypogonadism. Bisphosphonates, denosumab, iron chelation therapy, and hormone replacement therapy are effective in reducing bone resorption and improving bone health in thalassemia patients.

Emerging therapies targeting VEGF and RANKL signaling represent promising new approaches for treating bone resorption in thalassemia. Further research is needed to better understand the molecular mechanisms driving bone loss in thalassemia and to develop more effective therapeutic interventions for preventing long-term skeletal complications.

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