Review Article

HEPCIDIN AS A BIOCHEMICAL PARAMETER FOR THE ASSESSMENT OF IRON DEFICIENCY ANEMIA

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Abstract

Iron deficiency anemia is the most prevalent nutritional disorder in the world. Information on the metabolism of hepcidin and its possible significance as a biochemical parameter in iron deficiency anemia is reported in this review, which was based on a survey of the databases PubMed and LILACS for articles published between 2006 and 2010 on hepcidin as a biomarker for the regulation of iron metabolism. The literature search returned 35 studies published in international journals and one study on the subject in a Brazilian journal. Hepcidin production is homeostatically regulated by anemia and hypoxia. When oxygen delivery is inadequate, hepcidin levels decrease. Consequently, more iron is made available from the diet and from the storage pool in macrophages and hepatocytes. Hepcidin binds to ferroportin, regulating iron release into plasma. When hepcidin concentrations are low, ferroportin molecules are displayed on the plasma membrane and release iron. When hepcidin levels increase, hepcidin binds to ferroportin molecules inducing their internalization and degradation, and iron release is decreased progressively. Apparently, the development of diagnosis and therapy for anemia based on the bioindicator hepcidin may provide a more effective approach. Epidemiological studies are needed to demonstrate the relevance of hepcidin to the differential diagnosis of anemia, including sampling protocols for analysis, with standardization similar to that used in other biochemical assessments, and establishment of cutoff points for urinary and plasma expression of this peptide.

Key words: Anemia. Iron. Parameters.

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Introduction

Hepcidin and iron metabolism

Iron is an essential element for nearly all living organisms. Iron is a key component of oxygen storage and transporting proteins, such as hemoglobin and myoglobin, and of many enzymes that catalyze oxidation-reduction reactions necessary to generate energy and produce various metabolic intermediates for host defense[1-3].

In all species, the concentration of iron in biological fluids is tightly regulated to provide iron as needed and to avoid toxicity, since iron excess can lead to the generation of reactive oxygen species (ROS), and decreased iron levels can lead to anemia[4,5]. Thus, maintenance of body iron stores is essential, because many human diets contain iron sufficient only to replace the small iron losses. When iron intake is more abundant, apparently iron absorption is appropriately controlled [6,7].

Iron deficiency may occur by inadequate dietary intake, by increased physiological needs of the nutrient, and/or by increased losses, which may lead to anemia

Anemia is the most widespread nutritional disorder in the world, affecting mainly women of childbearing age and children under two years of age [8]. The same scenario is described in Brazilian studies [9-12]. Data from the last National Survey on Children and Women Health revealed that 20.9% of Brazilian children and 29.4% of women have anemia[13].

For this reason, there is great interest in investigating effective methods for the diagnosis of iron deficiency anemia.

Recent studies have evaluated the use of hepcidin as a biomarker for the regulation of iron metabolism. Hepcidin has evolved as the primary regulator of iron homeostasis and a probable mediator of anemia of chronic disease and inflammation. This role has been widely demonstrated in a number of recent studies[14-18], and there is enormous interest in quantifying circulating hepcidin levels in clinical samples[19].

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Objective

The objective of this study was to gather information on the metabolism of hepcidin and its possible significance as a biochemical parameter for the assessment of the amount of iron absorbed in individuals with iron deficiency anemia.

Methods

A review of the literature was conducted based on a survey of the databases PubMed (National Library of Medicine's Medline Biomedical Literature) and LILACS (Latin American and Caribbean Literature) for articles published between 2006 and 2009, using "hepcidin" as the subject heading (MeSH) and search descriptor, respectively. The literature search returned 501 articles on the subject; 500 studies published in international journals and one study in a Brazilian journal.

English- and Portuguese-language scientific publications that referred to hepcidin as a new biomarker for the regulation of iron metabolism were selected. Studies that related hepcidin to hemochromatosis, thalassemia, and anemia of chronic disease and inflammation were excluded, resulting in 36 studies: 35 articles published in international journals and one article in a Brazilian journal [20].

Hepcidin expression related to iron deficiency anemia

The discovery of hepcidin has given rise to a considerable number of studies on this topic. Hepcidin is a circulating peptide hormone composed of 25 amino acids, synthesized in the liver and detectable in blood and urine [3,21].

As the master regulator of systemic iron homeostasis, hepcidin coordinates the use and storage of this mineral in the body[4]. Hepcidin acts on the inhibition of intestinal iron absorption and iron release by macrophages and enterocytes[16,17], and is a mediator in the cycle of iron absorption between the liver and intestine [22-24].

Measurement of hepcidin concentrations can be used for different diagnoses of anemia, such as iron deficiency anemia, which is characterized by low levels of this hormone. In principle, hepcidin measurements could complement the most frequently used indicators of total body iron stores, such as serum iron and ferritin[2,25,26], in addition to others such as transferrin receptor, transferrin saturation, and zinc protoporphyrin[27].

Iron homeostasis is regulated by two main mechanisms: an intracellular mechanism, dependent on the amount of iron available for the cell, and a systemic mechanism, in which hepcidin plays a crucial role [20,28].

Most of the iron absorbed from the diet or recycled from hemoglobin is intended for developing erythrocytes, whose production is increased in response to erythropoietic stimuli, such as blood loss or hypoxia.

Hepcidin production is homeostatically regulated by anemia and hypoxia[2,29], in addition to being regulated by inflammation and oxidative stress [30-33].

When oxygen delivery is inadequate, the homeostatic response is an increased production of erythrocytes. Hepcidin levels then decrease, as well as its inhibitory effects. Consequently, more iron is made available from the diet and from the storage pool in macrophages and hepatocytes[2].

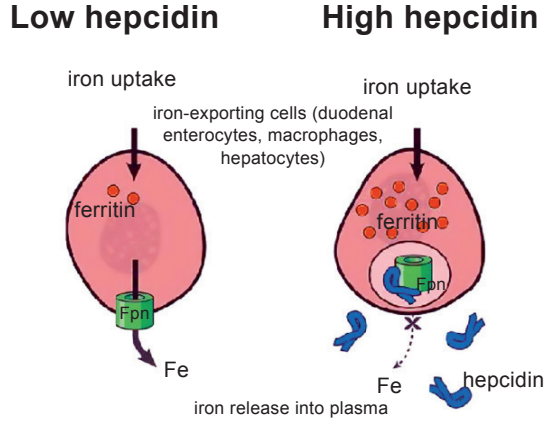
Depending on the cell type, iron can be taken up by distinct pathways. Bioavailable iron in the diet is mostly present in both the ferric form (Fe^3) and as heme. The uptake of Fe^3 is mediated by duodenal cytochrome b reductase enzyme – Dcytb, reducing Fe^3 to its ferrous form (Fe^2), which is carried across the cell membrane by divalent metal transporter 1 – DMT1 [2].

Inside the cell, absorbed Fe^2 binds to ferritin heading to the basolateral membrane, where ferroportin is located, present in enterocytes, macrophages, and hepatocytes, which is a protein required for Fe^2 transport into plasma[20,34-36].

Hepcidin binds to ferroportin, regulating iron export into plasma. When hepcidin concentrations are low, ferroportin molecules are displayed on the plasma membrane and export iron. When hepcidin concentrations increase, hepcidin binds to ferroportin molecules inducing their internalization and degradation, and iron release is decreased progressively [3,36] (Figure 1).

Therefore, in healthy patients, hepatic hepcidin production is regulated by a feedback mechanism induced by circulating iron. Low levels of circulating hepcidin allow ferroportin to release iron into the bloodstream; elevated levels of hepcidin effectively reduce iron absorption in enterocytes by disabling the iron exporter ferroportin [37,38]. Apparently, this mechanism observed in ferroportin present in enterocytes functions differently from ferroportin of macrophages or hepatocytes [34,39,40].

Figure 1 - Mechanism of hepcidin-mediated cellular iron regulation



Fpn = ferroportina (adaptado de Ganz [3]).

Anemia induces a cascade of changes that individually or in combination suppress hepcidin expression. The decrease or absence of urinary hepcidin is the expected response in patients with iron deficiency anemia, such as patients with anemia caused by gastrointestinal blood loss. A study by Pak et al.[41] found that anemia does not directly regulate hepcidin expression, but exerts its effects on this hormone and iron metabolism through an as yet uncharacterized substance released during erythropoiesis. The specific mediators and pathways by

which these elements influence hepcidin synthesis and release remain to be elucidated.

The diagnosis of iron deficiency anemia within the context of anemia of chronic disease is commonly performed with routine biochemical parameters, such as transferrin saturation, C-reactive protein, and, less often, transferrin receptor and zinc protoporphyrin. Each has their own disadvantages. In contrast to the increased levels of hepcidin in anemia of chronic disease, both in vitro and the classic iron deficiency anemia in humans are associated with low hepcidin expression, which makes this hormone a potential marker for detection of iron deficiency anemia coexisting with anemia of chronic disease [42].

Anemia related to iron levels in humans and animal models highlights the importance of erythropoiesis in the regulation of hepcidin and the need to understand its molecular basis. Moreover, the development of diagnosis and therapy for anemia based on hepcidin may provide a more effective approach to prevent toxicity associated with iron overload [43].

Relationship of hepcidin with other molecules and biomarkers of iron nutritional status

Hepcidin concentration is apparently regulated by molecules such as transferrin receptor 2 – TfR2, HFE (product of a high-iron gene), and hemojuvelin – HJV. Studies in hepatocytes show that hepcidin responds to iron only when it is bound to transferrin, suggesting that the liver uses transferrin saturation as an iron sensor. At the cellular level, iron sensing appears to involve TfR2 and HFE. Patients with mutations in HFE do not produce hepcidin in response to oral iron as do normal individuals. In addition, other proteins are involved in hepcidin expression, including bone morphogenic proteins, among others [37]

Conclusion

Recent studies have contributed to our knowledge of the major regulatory pathways of iron homeostasis in the body, highlighting the role that hepcidin plays in this process. Measurement of this hormone has the potential to become an important tool for diagnosis and treatment of anemia and diseases caused by iron metabolism disorders.

The studies published so far are based on clinical trials, consisting of a limited number of cases. Epidemiological studies are needed to demonstrate the role of hepcidin in the differential diagnosis of anemia, including sampling protocols for analysis of hepcidin with standardization similar to that used in other biochemical assessments and establishment of cutoff points for urinary and plasma expression of this peptide.

A better understanding of the precise interaction between ferroportin and hepcidin might pave the way for the development of new drugs that can alter hepcidin activity.

Furthermore, the development of accessible and accurate laboratory tests is important to assess alterations in hepcidin expression in various populations. Although some methods for the production of recombinant hepcidin have been reported, further research is needed to provide pure hepcidin for the development of clinical trials in humans.

Conflict of interest: No conflicts of interest declared concerning the publication of this article.

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Artigo recebido: 24/02/10
Aceito para publicação: 04/05/10