Borneo Journal of Pharmacy
Vol 6 Issue 4 November 2023
Pages 360 – 369

https://journal.umpr.ac.id/index.php/bjop/article/view/4411
DOI: https://doi.org/10.33084/bjop.v6i4.4411
e-ISSN: 2621-4814

Mini Review

Iron-Overload Conditions: Manifestations to the Kidney Organs – A Review

Nadia Larasinta Heriatmo [1,2]

Ari Estuningtyas [3*]

Vivian Soetikno [3]

[1] Master Program in Biomedical Sciences, Universitas Indonesia, Central Jakarta, Special Capital Region of Jakarta, Indonesia

[2] Department of Pharmacy, Universitas Pancasila, South Jakarta, Special Capital Region of Jakarta, Indonesia

[3] Department of Pharmacology and Therapeutics, Universitas Indonesia, Central Jakarta, Special Capital Region of Jakarta, Indonesia

*email: ari_handoyo@yahoo.com

Keywords:
Ferritin
Hemosiderosis
Iron deposits
ROS
Transfusion

Abstract

Excess iron is a risk factor for organ dysfunction and damage resulting in various organ diseases such as liver, heart, and kidney, diabetes mellitus, and neurodegenerative diseases. Iron overload in some individuals is caused by various factors, including genetic predisposition such as genetic hemochromatosis, repeated transfusion of red blood cells, and parenteral iron administration in conditions of transfusion-dependent anemia. A disturbance in the globin gene in diseases such as β-thalassemia major causes an imbalance of the globin chain, resulting in chronic anemia in the sufferer. It has been reported that the human body does not have a mechanism for eliminating excess iron levels. Routine transfusion has become a solution to overcome chronic anemia so that patients can maintain hemoglobin levels, and the result of this transfusion repetition is the accumulation of iron in various organs, such as the heart, liver, endocrine glands, pancreas, lungs, and kidneys. Excess iron can be toxic to the body due to the formation of harmful free radicals that can damage cells and tissues. An increase in excessive ROS can result in the saturation of the antioxidant system. The presence of free radicals can lead to damage and the occurrence of filtration dysfunction in the glomerulus.

Received: December 13th, 2022
1st Revised: May 10th, 2023
2nd Revised: September 26th, 2023
Accepted: November 27th, 2023
Published: November 30th, 2023

INTRODUCTION

Iron overload is when too much iron accumulates in the body. Increased iron absorption in the small intestinal mucosa and frequent red blood cell transfusions can lead to iron overload in patients with chronic anemia such as β-thalassemia major. Before recently, long-term iron chelation therapy with iron chelating medications was the standard for managing iron overload, particularly in patients with transfusion-dependent thalassemia[1]. Hemosiderosis is an iron overload condition with increased iron deposition in tissues without clinical signs[2]. More than 3% of people worldwide carry the thalassemia gene, with Southeast Asia having the highest incidence – up to 40% – of this single gene inherited disease. Thalassemia is an inherited disorder of globin chain synthesis. Various mutations and deletions in the α- and β-globin gene clusters reduce globin chain synthesis to varying degrees and produce thalassemia traits or clinical features of thalassemia. Chronic anemia brought on by inefficient erythropoiesis, intra and extramedullary hemolysis, and iron overload dominates the clinical picture of thalassemia[3]. Iron is essential for transporting oxygen and various metabolic processes. Its ability to take and donate electrons, switching between its ferrous (Fe^{2+}) and ferric (Fe^{3+}) forms, enables it to participate in this process[4]. Hereditary anemia caused by thalassemia is typically treated with routine blood transfusions, which may raise the body's overall iron levels. As soon as proteins that control iron are functional (e.g., ferritin, transferrin)[5]. Based on the severity of the clinical phenotype, β-thalassemia has historically been categorized into three primary subgroups: major, intermedia, and minor. Red blood cell transfusion requirements have been used as the basis for a more straightforward and clinically

How to cite: Heriatmo NL, Estuningtyas A, Soetikno V. Iron-Overload Conditions: Manifestations to the Kidney Organs – A Review. Borneo J Pharm. 2023;6(4):360-9. doi:10.33084/bjop.v6i4.4411

applicable classification system over the past ten years. Thalassemia patients are classified as transfusion-dependent (TD) or non-transfusion-dependent (NTD). While NTD patients frequently have anemia, they do not require blood transfusions for daily function or survival; TD thalassemia patients must receive regular blood transfusions throughout their lifetimes. Patients with NTD thalassemia may occasionally need transfusions, notably during pregnancy, surgery, and infections, but they typically maintain hemoglobin (Hb) concentrations between 7 and 10 g/dL[6]. Reactive oxygen species (ROS) and free radicals, which harm cellular and subcellular structures and result in metabolic dysfunction, are produced by metals like iron and some low molecular weight substances containing iron. The presence of disturbed iron metabolism can also produce free radicals[3]. Iron deposits in essential organs like the kidneys, liver, and heart may occur from circulating non-transferrin-bound iron (NTBI), brought on by plasma transferrin saturation and iron overload[7]. Iron chelating agents must be supplied continually and for the rest of one's life to remove excess iron from the body and stop it from building up in tissues to combat the accumulation of too much iron (iron overload)[8]. Renal illness, like in other aging populations, potentially may become more prevalent with the arrival of efficient chelating medicines that can decrease the iron burden and its effects and extend patients' survival[9].

Iron overload is brought on by a buildup of iron that starts in the blood plasma and eventually gets deposited in tissues and organs, causing organ damage, especially in patients receiving routine blood transfusions. Kidneys, as one of the vital organs involved in excretion, might be affected by their function if damaged. There has yet to be much discussion on how iron overload affects kidney damage, how it manifests, or how it relates to renal function damage markers. How iron can harm the kidney will be discussed in further detail.

THE ROLE AND FUNCTION OF THE KIDNEYS

The kidneys play a critical role in maintaining homeostasis by controlling the amounts of various plasma elements, including electrolytes and water, and by eliminating all metabolic wastes other than CO_2 exhaled by the lungs. Plasma will be repeatedly filtered through the kidneys, retaining the constituents of value to the body and removing unwanted or excessive materials into the urine. Controlling the salt and water balance allows the kidneys to regulate the volume and osmolarity of the solute concentration of the internal fluid environment[10]. Besides playing a crucial regulatory role in preserving fluid and electrolyte balance, the kidneys are vital for the body's excretion of potentially hazardous metabolic waste products and foreign substances. These wastes must be eliminated in solution since they cannot be eliminated as solids. As a result, the kidneys must produce at least 500 mL of waste-containing urine per day. **Figure 1** shows the locations of the three fundamental procedures that contribute to the creation of urine: glomerular filtration, tubular reabsorption, and tubular secretion[11]. The kidneys are the primary excretory organs for eliminating metabolic waste products and are crucial for preserving the body's normal electrolyte and fluid balance. Additionally, the kidneys perform two endocrine tasks: they secrete erythropoietin, which encourages the creation of red blood cells, and renin, which controls blood pressure[12].

IRON HOMEOSTASIS

Plasma iron is quantitatively much less than erythrocytic iron and has a total mass of around 5 mg, and erythrocytic iron, which makes up roughly half of the body's iron stores, is the two main types of blood iron that circulate. Erythrocytic iron is the constituent of heme incorporated into the four globin molecules to create the hemoglobin macromolecule. Furthermore, plasma iron must be attached to transferrin since it exists as a metal and cannot flow freely in a liquid. A transferrin molecule can only bind a maximum of two iron atoms[13]. The primary sources of iron are duodenal enterocytes and recycled macrophages. Absorbed iron is transported across the apical membrane of enterocytes after reduction. Imported iron can be used for cell-intrinsic metabolism, stored bound to the protein ferritin, or exported to plasma bound to the protein transferrin[14]. Dietary iron is present in the oxidized form Fe^{3+} and must be reduced to Fe^{2+} before absorption occurs in the intestine. This reduction is thought to be mediated by ferrireductase in the apical membrane of intestinal cells[15]. Cytochrome b (Dcytb) in the duodenal enterocyte membrane reduces Fe^{3+} to Fe^{2+}, then Fe^{2+} enters the cell through divalent metal transporter 1 (DMT1) on the membrane. The protein ferritin, which has enormous cavities to store hundreds of iron atoms and is coupled to extracellular iron, resists the dissociation of iron and results in oxidative cell damage[16]. Under typical

circumstances, transferrin-bound iron makes up most of the iron in the bloodstream. Iron is kept in an inert form by transferrin, which may carry up to two iron molecules[15]. Intracellular iron is released into the circulation via the only known iron exporter, ferroportin, commonly referred to as FPN1, and encoded by the *SLC40A1* gene[16,17]. After Fe^{2+} comes out into the circulation, it will be oxidized to Fe^{3+} by ferroxidases such as hephaestin (HEPH) or its homolog: ceruloplasmin (CP) and will then be bound to transferrin protein/transferrin bound iron (TBI) and transported by the bloodstream[16]. Specific uptake mechanisms in the body transfer circulating iron to erythrocytes and other cells. The main mechanism is transferrin-bound iron uptake by TFR1 via receptor-mediated endocytosis into the clathrin-coated (**Figure 2**). Iron will be released in an acidic environment from the endosome, then Fe^{3+} is reduced to Fe^{2+} by the STEAP3 ferrireductase, and Fe^{2+} is exported out of the endosome by DMT1, while TFR1 will be recycled to the cell surface[15].

Hepcidin, an iron hormone released by the liver, is the mechanism by which systemic iron is regulated under physiologically normal circumstances. Hepcidin binds to ferroportin (a transmembrane iron-exporting protein) on macrophages and iron-storing hepatocytes, degrading ferroportin and inhibiting iron entry into the circulation. Moreover, hepcidin binds to ferroportin on enterocytes, reducing the expression of DMT-1 protein that facilitates non-heme iron uptake on the apical surface of enterocytes and decreasing intestinal iron absorption[18]. Mutations that cause a lack of hepcidin or resistance to it cause excessive amounts of iron to be absorbed from the duodenum, released uncontrollably into the circulation by macrophages, and deposited excessively in several organs[19]. Iron is necessary for healthy physiology, but too much of it can be dangerous because it speeds up the Fenton reaction, which creates reactive oxygen species (ROS) that can destroy cells and tissues. Maintaining body iron homeostasis is critical because there is no natural method to eliminate excess iron from the body[15]. Iron overload results in plasma transferrin saturation and hence the emergence of circulating non-transferrin-bound iron (NTBI), which causes iron deposition in vital organs such as the kidneys, liver, and heart[7,19].

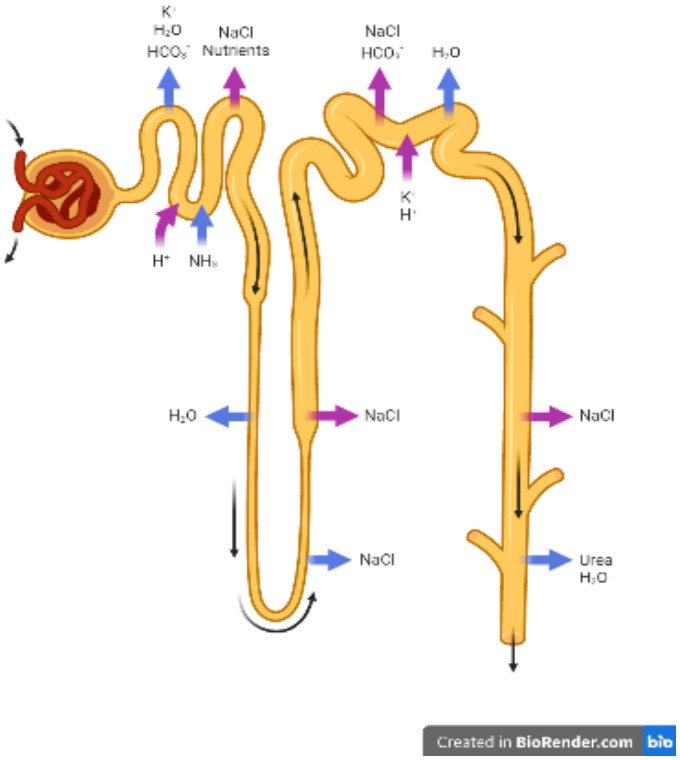

Figure 1. Kidney nephron, created by https://www.biorender.com/. The fundamental mechanism in the renal system results in urine generation. Glomerular filtration: nondiscriminant filtration of protein-free plasma from the glomerulus into Bowman's capsule. Tubular reabsorption: selective movement of filtered substances from the tubular into the peritubular capillaries. Tubular secretion: selective movement of nonfiltered substances from the peritubular capillaries into the tubular lumen[10].

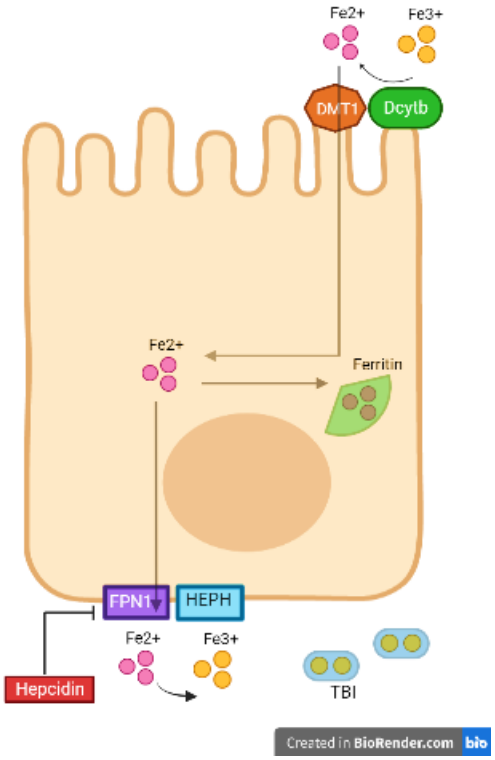

Figure 2. Iron homeostasis, created by https://www.biorender.com/. Non-heme iron is mainly absorbed by intestinal enterocytes expressing the ferric reductase DCYTB, which converts Fe^{3+} to Fe^{2+} and facilitates its transport through the apical side of the enterocyte cell membrane by DMT1 subsequently stored in cells as bound Ferritin protein. Iron enters the circulation by being exported through FPN on the basolateral membrane of enterocytes. Next, the formed TBI complex[18].

IRON OVERLOAD CONDITIONS

β-thalassemia is a hereditary disorder of Hb production characterized by impaired Hb synthesis, leading to shorter red blood cell survival through hemolysis and premature death of red blood cell precursors in the bone marrow. This condition results in chronic severe anemia and bone marrow expansion. Hepatosplenomegaly, bone deformities brought on by bone marrow growth, and heart failure brought on by severe anemia can all result from thalassemia if left untreated[20]. Red blood cell transfusions are essential for the survival of patients with β-thalassemia. Pediatric thalassemia patients require erythrocyte transfusions more frequently than 8–12 times per year in order to ensure their survival as well as healthy growth and development[21]. Blood transfusions help treat chronic anemia, avoid bone deformities, stimulate healthy growth, and improve patients' quality of life. For adult men and women, Hb levels exceeding 120 and 130 g/L, respectively, are considered normal values[20]. Due to repeated transfusions, the body cannot store enough iron to reach acceptable levels of excess iron in the plasma. By producing hydroxyl free radicals, which lead to oxidative stress, excessive iron has the potential to penetrate cells in the liver, heart, endocrine glands, and other organs and gradually destroy surrounding tissue. By binding plasma iron to transferrin, which becomes highly saturated in individuals with iron overload, the body can only manage iron to a certain extent. This causes the production of a significant amount of iron not linked to transferrin, including labile plasma iron, and speeds up the creation of free radicals in some organs and tissues. Apart from transferrin, the human body itself does not have any physiological mechanisms to remove excess iron[22].

PATHOPHYSIOLOGY OF KIDNEY DAMAGE DUE TO IRON OVERLOAD

Urine formation is regulated by a highly sophisticated mechanism involving the glomerulus, proximal convoluted tubule, loop of Henle, and distal convoluted tubule. The wound's severity and location can significantly impact the outcomes of any incident that damages the tubular epithelial cells. According to experimental research, mice with an excess of iron can develop mild proteinuria, considerable glomerulosclerosis, tubular atrophy, interstitial fibrosis, and visible iron deposits in

the proximal tubules, glomeruli, and interstitium[23]. Chronic renal hyperfiltration causes increased proteinuria and, in the long term, can lead to a progressive decrease in glomerular filtration rate (GFR) and renal fibrosis. The reasons for the nephrolithiasis seen in TDT patients have been hypothesized to be hemolysis, hypercalciuria, and hyperuricosuria. In 2 to 10% of TDT patients, hematuria has been noted, and it may be linked to an increased risk of nephrolithiasis[24]. Almost every living thing on Earth depends on iron, the second most abundant metal in the world. Humans require iron as a cofactor for hemoproteins (catalase, cytochrome, hemoglobin, or myoglobin) and other non-heme proteins involved in cell division, proliferation, DNA synthesis, and drug metabolism. On the other hand, through the Fenton reaction, iron can also create dangerous oxygen-free radicals, specifically hydroxyl radicals. Normal cellular metabolism produces a slight excess of free radicals, which are then neutralized by natural cellular antioxidant systems. However, in conditions of excess iron, such as hemochromatosis, the level of non-transferrin bound iron (NTBI), i.e., free iron circulating in the plasma, will increase in number. The availability of free iron will form the basis of iron toxicity because it accelerates the Fenton reaction to produce an increase in the amount of ROS that cannot be suppressed so that saturation of the antioxidant system can occur[19].

A condition of imbalance between excessive oxidants, also known as free radicals, and the antioxidant system's insufficient ability to degrade these radicals as a kind of defense against them is known as oxidative stress. Reactive oxygen and reactive nitrogen species (RNS) are two examples of oxidants created and eliminated by various antioxidant defense systems during physiological conditions[25]. Reactive oxygen species play a significant role in cell signaling involved in cell proliferation, differentiation, apoptosis, and immune defense in various cells, including kidney cells, under normal settings. Numerous inflammatory cells are attracted by the creation of ROS, which also promotes the production of inflammatory cytokines, growth factors, and transcription factors. Reactive oxygen species and RNS levels within cells are enhanced during oxidative stress, frequently seen in several kidney illnesses. Reactive oxygen species are typically considered hazardous substances, resulting in oxidative damage since they interact with lipids, proteins, and DNA. Both the renal cortex and the medulla can experience ROS generation or altered ROS production, which can have a variety of repercussions, from altered renal blood flow to sodium/fluid retention to inflammation and cellular fibrosis, as well as the onset of proteinuria[26].

In the course of renal failure, it has been reported that high levels of free radicals are associated with disease pathogenesis and progression[27]. Albumin can be found in the urine of patients with glomerular or tubular renal injury, but higher urinary albumin levels are usually found in glomerular injury[28]. Albuminuria is a recognized indicator of renal impairment that develops early in many types of chronic kidney disease (CKD) and plays a significant role in the course of the illness by causing mesangial and tubular toxicity as well as activating systemic inflammatory pathways[29]. The occurrence of barrier dysfunction in glomerular filtration in the presence of podocyte injury is a sign of the development of proteinuria and subsequent glomerulosclerosis. Evidence from experimental and clinical studies suggests podocytes are particularly vulnerable to oxidative harm. It appears that glomerular injury is directly mediated by an increase in ROS in an experimental model of the disease that develops into focal segmental glomerulosclerosis (FSGS)[30].

MARKERS OF IRON OVERLOAD AND KIDNEY DAMAGE

Ferritin and transferrin saturation

A significant protein that stores iron, ferritin is also involved in several physiological and pathological processes and is crucial for maintaining iron homeostasis. Its primary function is as a ferroxidase in iron absorption, converting Fe (II) to Fe (III). Iron is hazardous to cellular systems because it can generate reactive species that can obliterate DNA and proteins. Iron pools within cells are supported and captured by ferritin. Ferritin is mainly utilized in clinical medicine as a serum marker of total body iron reserves. Serum ferritin is crucial for diagnosing and treating both iron deficiency and excess[31]. The ratio of serum iron to total iron binding capacity, or transferrin saturation (TSAT), is another significant biochemical marker for determining the body's overall iron status. In CKD, transferrin saturation can track a child's response to iron therapy and ESAs (erythropoiesis-stimulating drugs). Iron disutilization for erythropoiesis is thought to occur in patients with high ferritin and TSAT levels. Even if the body appears to have adequate iron stores, iron disutilization for erythropoiesis occurs when insufficient iron is incorporated into erythroid precursors. Patients with infectious diseases, persistent inflammation, chronic heart disease, chronic kidney disease, and malignant diseases are affected by this condition[32]. According to the Kidney Disease Outcomes Quality Initiative (K/DOQI), serum ferritin and transferrin saturation were the primary markers

in evaluating iron management in patients with anemia and CKD. According to the K/DOQI guidelines, patients with serum ferritin levels greater than 800 ng/mL may experience iron overload, albeit this condition is also very variable. In patients undergoing dialysis, serum ferritin levels reach 2000 ng/mL, which is a picture of the condition of patients with hemochromatosis, where the deposition of iron in the tissue clinically begins to occur[33].

Urea

The liver produces and distributes urea, the byproduct of protein and amino acid catabolism, through internal and extracellular fluids, which are then filtered in the glomerulus. Urea examination is beneficial in establishing the diagnosis of acute renal failure. The clinical index most often determined to estimate renal function depends on the serum urea concentration. An increase in Blood Urea Nitrogen (BUN) is linked to the presence of kidney failure and obstruction of the urinary tract by kidney stones. Urea clearance is a measure of abnormalities in the glomerular filtration rate. When the BUN value exceeds 100 mg/dL, it may be a sign of significant kidney damage[34].

Creatinine

Creatinine is a product of the breakdown of creatinine phosphate in muscles, and depending on the amount of muscle mass, the body typically produces it at a reasonably steady rate. Typically, creatinine is used to gauge renal function. Men's normal creatinine readings are 110-150 mL/min, whereas women's normal values are 100-130 mL/min. The progression of renal disease is tracked using measurements of creatinine clearance[35]. According to the National Kidney Disease Education Program of the National Institutes of Health, the estimated GFR should be determined using serum creatinine readings as an early indicator of kidney disease[36]. When the serum creatinine level is higher than the top limit of the normal range, the diagnosis will result in renal failure. The glomerulus and tubules eventually excrete less creatinine in patients with chronic renal failure and uremic diseases[34]. Based on the evidence from Ige *et al.*[36] showed that in experimental Wistar rats treated with ferrous sulfate treatment as much as 15 and 30 mg/kg BW every day for 21 days, the serum analysis showed an increase in urea and creatinine levels in both treatment groups significantly compared to the control group.

Renal tissue antioxidant

There have been reports of renal impairment occurring alongside oxidative tissue damage when there is inflammation[37]. Chronic kidney disease's pathophysiological foundation includes OxS, apoptosis, and inflammation. These characteristics are consistently present in both humans and animals. They act similarly in rat models of chronic renal failure, making them essential disease mediators. High plasma concentrations of inflammatory mediators (C-reactive protein, tumor necrosis factor-α (TNF-α), and other cytokines) and many stress markers are present in patients and laboratory animals with CKD[38]. Early reactions to kidney injury include activating inflammatory pathways inside the kidneys and recruiting inflammatory cells to the area of injury. Injury may be followed by healing mechanisms or chronic inflammation that progresses over time and eventually results in fibrosis[39]. The first line of defense against free radicals or ROS/RNS produced by enzymatic reactions or auto-oxidation mechanisms in mammalian systems is antioxidants. The measurement of total antioxidant status (TAS), total oxidant status (TOS), and oxidative stress index (OSI) in plasma or body tissues provides the cumulative action of all the antioxidants and oxidants present in the body[40]. An imbalance between the production of free radicals and the antioxidant system results in oxidative stress (OxS), which harms biomolecules. These antioxidant systems are vital in preserving the cell's oxide/ reduction equilibrium[41]. Polyunsaturated fatty acids in membrane phospholipids are the primary target substrates for free oxygen radical activity, and their alteration leads to the disruption of cell structure and function. The end product of these reactions is malondialdehyde (MDA). It is excreted in urine, blood, and other fluids and, therefore, serves as a marker of lipid peroxidation and the presence of oxidative stress, respectively. Reactive oxygen species must be deactivated and removed to the activation of antioxidant defense mechanisms, such as the following enzymes superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPX)[42].

Proinflammatory mediators

Tumor necrosis factor-α, interleukin 1 (IL-1), IL-6, IL-8, IL-12, monocyte chemoattractant protein 1, tissue inhibitor of metalloproteinase 1, and interferon (IF) are a few examples of chemokines and cytokines that have been found to have

higher levels of proinflammatory mediators. These mediators are crucial in the inflammatory process that harms organs[37]. Interleukin 6 influences the dynamics of the extracellular matrix, general renal hypertrophy, thickening of the basement membrane in the glomeruli, podocyte hypertrophy, and cell cycle arrest, all linked to albuminuria. Interleukin 6 also contributes to neutrophil infiltration in the interstitial tubules[43]. Compared to other proinflammatory mediators in the inflammatory response during acute renal impairment, it has been shown that the major proinflammatory mediator, IL-6, is a better marker in renal impairment patients[39]. Both glomerular and tubular cells displayed elevated TNF mRNA expression in the early stages of diabetes. Tumor necrosis factor-causes various effects, including the production and differentiation of inflammatory cells, renal cell cytotoxicity, apoptosis activation, glomerular hemodynamics modifications, increased vascular endothelial permeability, and increased oxidative stress. Blood cells and numerous intrinsic renal cells, including glomerular, endothelial, tubular, and mesangial cells, can produce inflammatory cytokines in the kidney. As nephropathy worsens, these chemicals' concentrations rise, and there is no connection between these inflammatory markers and urinary albumin excretion[43]. Proinflammatory cytokines, including IL-1, IL-6, and TNF-α, produce changes in renal structural hemodynamics, cellular necrosis, and alterations in the kidney's glomerular endothelial permeability[44]. Based on research conducted by Ige *et al*.[36], which showed that in experimental animals, Wistar rats treated with ferrous sulfate treatment of 15 and 30 mg/kg BW every day for 21 days showed that in serum analysis, there was an increase in TNF-α and IL-6 levels in both groups were significantly compared with the control animal group.

Finally, people with thalassemia who require blood transfusions may develop an iron overload disease that may result in organ damage. The condition of excess iron occurs due to the accumulation of iron in various organs. The accumulation that forms in the vital organs of the kidneys can cause oxidative stress, which can harm cells and organs, with the kidney being one of the most susceptible organs. Damage can occur in any area of the kidney, beginning with the glomerulus and progressing to the proximal tubule, the loop of Henle, and the distal tubule. As the kidney's primary function, it will impact every step of the excretion process. Being a crucial organ that also serves as an excretory organ, it is only reasonable that iron overload would substantially impact it if its function decreased and worsened. This happens because physiologically, excess iron cannot be expelled in its whole; it must be in the form of a chelate.

CONCLUSION

Ultimately, iron overload can disrupt iron homeostasis, increase NTBI, and increase free radical generation, all of which contribute to and induce kidney organ dysfunction. Appropriate treatment is needed to prevent iron accumulation in the organs: iron chelation drugs. Iron chelation improves the quality of life of individuals who receive routine blood transfusions. Indonesia, one of the countries with abundant botanical species, has become an opportunity for drug development research on iron chelation therapies based on natural ingredients. Natural product-based treatments are currently being developed as complementary therapies to the primary treatment to help patients maximize the treatment outcomes.

ACKNOWLEDGMENT

None.

AUTHORS' CONTRIBUTION

Nadia Larasinta Heriatmo: collected data, writing the paper. **Ari Estuningtyas**: conceived and design the analysis, writing the paper. **Vivian Soetikno**: conceived and design the analysis, collected data.

DATA AVAILABILITY

None.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

1. Verna FD, Estuningtyas A. Hematological Profile of Iron Overload in Rats Administered with Fruit Extract of Mahkota Dewa (Phaleria macrocarpa). J Farmasi Galenika Galenika J Pharm. 2022;8(2):117–23. doi:10.22487/j24428744.2022.v8.i2.15936

2. Udani K, Chris-Olaiya A, Ohadugha C, Malik A, Sansbury J, Paari D. Cardiovascular manifestations in hospitalized patients with hemochromatosis in the United States. Int J Cardiol. 2021;342:117-24. doi:10.1016/j.ijcard.2021.07.060

3. Laksmitawati DR, Handayani S, Udyaningsih-Freisleben SK, Kurniati V, Adhiyanto C, Hidayat J, et al. Iron status and oxidative stress in β-thalassemia patients in Jakarta. Biofactors. 2003;19(1–2):53–62. doi:10.1002/biof.5520190107

4. de Lima TG, Benevides FLN, Filho FLE, Farias IS, Dourado DXC, Fontenele EGP, et al. Treatment of iron overload syndrome: A general review. Rev Assoc Med Bras. 2019;65(9):1216–22. doi:10.1590/1806-9282.65.9.1216

5. Estuningtyas A, Wahyuni T, Wahidiyat PA, Poerwaningsih EH, Freisleben HJ. Mangiferin and mangiferin-containing leaf extract from Mangifera foetida L for therapeutic attenuation of experimentally induced iron overload in a rat model. J Herbmed Pharmacol. 2019;8(1):21–7. doi:10.15171/jhp.2019.04

6. Taher AT, Cappellini MD. Luspatercept for β-thalassemia: beyond red blood cell transfusions. Expert Opin Biol Ther. 2021;21(11):1363-71. doi:10.1080/14712598.2021.1968825

7. Sumneang N, Siri-Angkul N, Kumfu S, Chattipakorn SC, Chattipakorn N. The effects of iron overload on mitochondrial function, mitochondrial dynamics, and ferroptosis in cardiomyocytes. Arch Biochem Biophys. 2020;680:108241. doi:10.1016/j.abb.2019.108241

8. Estuningtyas A, Setiabudy R, Wahidiyat PA, Freisleben HJ. The Role of Mangiferin in the Prevention of Experimentally Induced Iron Overload in an Animal Model. Drug Res. 2019;69(4):234–40. doi:10.1055/a-0667-8530

9. Bhandari S, Galanello R. Renal aspects of thalassaemia a changing paradigm. Eur J Haematol. 2012;89(3):187–97. doi:10.1111/j.1600-0609.2012.01819.x

10. Ashoor IF, Somers MJG. Physiology of the Developing Kidney: Fluid and Electrolyte Homeostasis and Therapy of Basic Disorders (Na/H2O/K/Acid Base). In: Avner E, Harmon W, Niaudet P, Yoshikawa N, Emma F, Goldstein S, editors. Pediatric Nephrology.Berlin, Heidelberg: Springer; 2015. doi:10.1007/978-3-642-27843-3_12-1

11. Sherwood L. Human physiology : from cells to systems. 7th edition. Belmont, CA: Brooks/Cole; 2013. p. 148–162.

12. Radi ZA. Kidney Pathophysiology, Toxicology, and Drug-Induced Injury in Drug Development. Int J Toxicol. 2019;38(3):215-27. doi:10.1177/1091581819831701

13. Brissot P, Troadec MB, Loréal O, Brissot E. Pathophysiology and classification of iron overload diseases; update 2018. Transfus Clin Biol. 2019;26(1):80–8. doi:10.1016/j.tracli.2018.08.006

14. Franke GN, Kubasch AS, Cross M, Vucinic V, Platzbecker U. Iron overload and its impact on outcome of patients with hematological diseases. Mol Aspects Med. 2020;75:100868. doi:10.1016/j.mam.2020.100868

15. Dev S, Babitt JL. Overview of iron metabolism in health and disease. Hemodial Int. 2017;21(Suppl 1):S6–20. doi:10.1111/hdi.12542

16. Li Y, Huang X, Wang J, Huang R, Wan D, Yang G. Regulation of Iron Homeostasis and Related Diseases. Mediators Inflamm. 2020;2020:6062094. doi:10.1155/2020/6062094

17. Sangkhae V, Nemeth E. Regulation of the iron homeostatic hormone hepcidin. Adv Nutr. 2017;8(1):126–36. doi:10.3945/an.116.013961

18. Zeidan RS, Han SM, Leeuwenburgh C, Xiao R. Iron homeostasis and organismal aging. Ageing Res Rev. 2021;72:101510. doi:10.1016/j.arr.2021.101510

19. Mehta KJ, Je Farnaud S, Sharp PA. Iron and liver fibrosis: Mechanistic and clinical aspects. World J Gastroenterol. 2019;25(5):521–38. doi:10.3748/wjg.v25.i5.521

20. Shah FT, Sayani F, Trompeter S, Drasar E, Piga A. Challenges of blood transfusions in β-thalassemia. Blood Rev. 2019;37:100588. doi:10.1016/j.blre.2019.100588

21. Cakan P, Yildiz S, Akyay A, Öncül Y. Erythrocyte transfusion restored heart rate variability in children with thalassemia major. Transfus Apher Sci. 2021;60(4):103156. doi:10.1016/j.transci.2021.103156

22. Ho WL, Chung KP, Yang SS, Lu MY, Jou ST, Chang HH, et al. A pharmaco-economic evaluation of deferasirox for treating patients with iron overload caused by transfusion-dependent thalassemia in Taiwan. J Formos Med Assoc. 2013;112(4):221–9. doi:10.1016/j.jfma.2011.08.020

23. Ponticelli C, Musallam KM, Cianciulli P, Cappellini MD. Renal complications in transfusion-dependent beta thalassaemia. Blood Rev. 2010;24(6):239–44. doi:10.1016/j.blre.2010.08.004

24. Tanous O, Azulay Y, Halevy R, Dujovny T, Swartz N, Colodner R, et al. Renal function in β-thalassemia major patients treated with two different iron-chelation regimes. BMC Nephrol. 2021;22(1):418. doi:10.1186/s12882-021-02630-5

25. Daenen K, Andries A, Mekahli D, Van Schepdael A, Jouret F, Bammens B. Oxidative stress in chronic kidney disease. Pediatr Nephrol. 2019;34(6):975–91. doi:10.1007/s00467-018-4005-4

26. Jha JC, Banal C, Chow BSM, Cooper ME, Jandeleit-Dahm K. Diabetes and Kidney Disease: Role of Oxidative Stress. Antioxidants Redox Signal. 2016;25(12):657–84. doi:10.1089/ars.2016.6664

27. Libetta C, Sepe V, Esposito P, Galli F, Dal Canton A. Oxidative stress and inflammation: Implications in uremia and hemodialysis. Clin Biochem. 2011;44(14–15):1189–98. doi:10.1016/j.clinbiochem.2011.06.988

28. Griffin BR, Faubel S, Edelstein CL. Biomarkers of Drug-Induced Kidney Toxicity. Ther Drug Monit. 2019;41(2):213-26. doi:10.1097/ftd.0000000000000589

29. Stenvinkel P, Chertow GM, Devarajan P, Levin A, Andreoli SP, Bangalore S, et al. Chronic Inflammation in Chronic Kidney Disease Progression: Role of Nrf2. Kidney Int Rep. 2021;6(7):1775-87. doi:10.1016/j.ekir.2021.04.023

30. Duni A, Liakopoulos V, Roumeliotis S, Peschos D, Dounousi E. Oxidative stress in the pathogenesis and evolution of chronic kidney disease: Untangling ariadne's thread. Int J Mol Sci. 2019;20(15):3711. doi:10.3390/ijms20153711

31. Knovich MA, Storey JA, Coffman LG, Torti SV, Torti FM. Ferritin for the clinician. Blood Rev. 2009;23(3):95–104. doi:10.1016/j.blre.2008.08.001

32. Kuragano T, Joki N, Hase H, Kitamura K, Murata T, Fujimoto S, et al. Low transferrin saturation (TSAT) and high ferritin levels are significant predictors for cerebrovascular and cardiovascular disease and death in maintenance hemodialysis patients. PLoS One. 2020;15(9):e0236277. doi:10.1371/journal.pone.0236277

33. Wish JB. Assessing iron status: beyond serum ferritin and transferrin saturation. Clin J Am Soc Nephrol. 2006;1(Suppl 1):S4–8. doi:10.2215/cjn.01490506

34. Gowda S, Desai PB, Kulkarni SS, Hull V V, Math AAK, Vernekar SN. Markers of renal function tests. N Am J Med Sci. 2010;2(4):170–3.

35. Miller WG, Myers GL, Ashwood ER, Killeen AA, Wang E, Thienpont LM, et al. Creatinine measurement: State of the art in accuracy and interlaboratory harmonization. Arch Pathol Lab Med. 2005;129(3):297–304. doi:10.5858/2005-129-297-cmsota

36. Ige AO, Ongele FA, Adele BO, Emediong IE, Odetola AO, Adewoye EO. Pathophysiology of iron overload-induced renal injury and dysfunction: Roles of renal oxidative stress and systemic inflammatory mediators. Pathophysiology. 2019;26(2):175–80. doi:10.1016/j.pathophys.2019.03.002

37. Beheshti F, Norouzi F, Abareshi A, Khazaei M, Alikhani V. Nigella sativa Prevented Liver and Renal Tissue Damage in Lipopolysaccharide-Treated Rats. Saudi J Kidney Dis Transpl. 2018;29(3):554–66. doi:10.4103/1319-2442.235184

38. Ali BH, Al-Salam S, Al Suleimani Y, Al Kalbani J, Al Bahlani S, Ashique M, et al. Curcumin Ameliorates Kidney Function and Oxidative Stress in Experimental Chronic Kidney Disease. Basic Clin Pharmacol Toxicol. 2018;122(1):65–73. doi:10.1111/bcpt.12817

39. Zhang WR, Parikh CR. Biomarkers of Acute and Chronic Kidney Disease. Annu Rev Physiol. 2019;81:309–33. doi:10.1146/annurev-physiol-020518-114605

40. Verma PK, Raina R, Sultana M, Singh M, Kumar P. Total antioxidant and oxidant status of plasma and renal tissue of cisplatin-induced nephrotoxic rats: Protection by floral extracts of Calendula officinalis Linn. Ren Fail. 2016;38(1):142–50. doi:10.3109/0886022x.2015.1103585

41. Aguilar TAF, Navarro BCH, Pérez JAM. Endogenous Antioxidants: A Review of their Role in Oxidative Stress. In: Morales-Gonzalez JA, Morales-Gonzalez A, Madrigal-Santillan EO, editors. A Master Regulator of Oxidative Stress - The Transcription Factor Nrf2. London, UK: IntechOpen; 2016. doi:10.5772/65715

42. Todorova I, Simeonova G, Kyuchukova D, Dinev D, Gadjeva V. Reference values of oxidative stress parameters (MDA, SOD, CAT) in dogs and cats. Comp Clin Path. 2005;13(4):190–4. doi:10.1007/s00580-005-0547-5

43. Pérez-Morales RE, Del Pino MD, Valdivielso JM, Ortiz A, Mora-Fernández C, Navarro-González JF. Inflammation in diabetic kidney disease. Nephron. 2019;143(1):12–6. doi:10.1159/000493278

44. Guarda NS, Bollick YS, De Carvalho JAM, Premaor MO, Comim FV, Moresco RN. High serum uric acid is associated with tubular damage and kidney inflammation in patients with type 2 diabetes. Dis Markers. 2019;2019:6025804. doi:10.1155/2019/6025804