Journal of Molecular Pharmaceutics & Organic Process Research

Mini Review Open Access

Changes in Erythropoiesis in Type 3 Hemochromatosis Mouse Models

Ramos Sousa*

Department of Clinical and Biological Sciences, AOU San Luigi Gonzaga, University of Torino, California, USA

Abstract

Type 3 haemochromatosis is a rare inherited iron excess condition that results in organ dysfunction. Mutations in the transferrin receptor 2 gene, which codes for two major isoforms, cause HFE3. Tfr2 is a hepatic regulator of the iron inhibitor hepcidin. Tfr2 is an intracellular isoform of the protein that controls iron levels in reticuloendothelial cells. Tfr2 is also important in erythropoiesis, as recently revealed. Tfr2 deficiency causes macrocytosis with reduced reticulocyte number and higher haemoglobin levels, as well as an increase in adult BM erythropoiesis and splenic erythropoiesis. Tfr2 deficiency, on the other hand, results in enhanced and immature splenic erythropoiesis. Taken together, our findings support the involvement of Tfr2 in erythropoiesis regulation and Tfr2 in promoting iron availability for erythropoiesis.

Keywords: Haemochromatosis; Hepatic; Iron inhibitor; Erythropoiesis; Macrocytosis

Introduction

Type 3 hemochromatosis, also known as transferrin receptor 2 deficiency, is a rare genetic condition that causes iron overload in several organs, most notably the liver. While the effect of iron accumulation on hepatic function has been extensively researched, new evidence suggests that altered erythropoiesis, the process of producing red blood cells, plays a critical role in the pathophysiology of type 3 hemochromatosis. Mouse models have provided vital insights into the underlying mechanisms of this illness as well as potential therapy methods. The purpose of this study is to investigate the link between altered erythropoiesis and type 3 hemochromatosis in mice models, offering light on the molecular mechanisms implicated and the implications for future research [1].

Mutations in the transferrin receptor 2 gene, which codes for two main isoforms, Tfr2 alpha and Tfr2 beta, exhibit considerable similarity to the type 1 transferrin receptor. TFR2 gene expression is not directly regulated by iron, unlike TFR1, and TFR2 mRNA lacks iron sensitive regions. As a result, no IRE-dependent posttranscriptional regulation of protein levels exists . Several in vitro experiments have shown that Tfr2 can bind iron-loaded transferrin, but with a much lower affinity than Tfr1.

Furthermore, TS regulates the amounts of this protein in the plasma membrane, with enhanced stabilization in the presence of highly saturated transferrin. The TFR2 isoform, which is a Tran's membrane protein, is produced through transcription of the whole TFR2 gene. Tfr2 is a shorter variant that lacks the cytoplasmic and trans membrane domains, and it is currently uncertain whether its activity is intracellular or extracellular. These two isoforms' expression patterns are also highly distinct. Tfr2 is significantly expressed in the liver and erythroleukemic cell lines, whereas Tfr2 is primarily expressed in the liver, brain, heart, and splenic macrophages. The majority of TFR2 mutations impair the synthesis of both Tfr2 isoforms. Some mutations, however, solely affect the Tfr2 isoform, whilst others, such as M172K, eliminate the Tfr2 methionine beginning codon [2].

Mutations in the TfR2 gene, which encodes a protein involved in iron homeostasis, are the primary cause of type 3 hemochromatosis. TfR2 works as an iron sensor in the body, signaling the liver to adjust iron absorption. The body fails to regulate iron absorption adequately in the absence of functional TfR2, resulting in iron buildup in different tissues.

TfR2-deficient animals have defects in erythroid progenitor development, resulting in less mature red blood cell output. These differentiation problems have been linked to deregulated signaling pathways such as the transforming growth factor beta and bone morphogenetic protein pathways [3].

Iron use and storage are strictly regulated mechanisms within erythroid cells. TfR2 deficiency impairs iron metabolism in erythroid cells, resulting in faulty haemoglobin synthesis and poor erythrocyte maturation, according to research.

Inflammation can have an effect on erythropoiesis by changing iron management and erythroid progenitor differentiation. According to research, enhanced inflammatory signaling contributes to erythropoiesis dysregulation in type 3 hemochromatosis mouse models, aggravating the iron overload phenotype [4].

Understanding the link between decreased erythropoiesis and type 3 hemochromatosis in mouse models can help develop therapeutic methods. Targeting key molecular pathways involved in erythroid differentiation and iron metabolism may hold promise for treating iron overload in patients.

Strategies aiming at restoring appropriate iron utilisation and storage within erythroid cells may be effective in alleviating the aberrant erythropoiesis seen in type 3 hemochromatosis.

Manipulation of hepcidin levels or activity could be a promising therapeutic method for normalizing iron absorption and iron release from erythroid cells, hence reducing iron excess.

Targeting inflammatory signaling pathways may aid in the restoration of normal erythroid differentiation and iron management, alleviating the effects of type 3 hemochromatosis [5].

***Corresponding author:** Ramos Sousa, Department of Clinical and Biological Sciences, AOU San Luigi Gonzaga, University of Torino, California, USA, E-mail: ramos.sousa@gamil.com

Received: 01-May-2023, Manuscript No: jmpopr-23-100640, **Editor Assigned:** 04-May-2023, pre QC No: jmpopr-23-100640 (PQ), **Reviewed:** 18-May-2023, QC No: jmpopr-23-100640, **Revised:** 22-May-2023, Manuscript No: jmpopr-23-100640 (R), **Published:** 29-May-2023, DOI: 10.4172/2329-9053.1000174

Citation: Sousa R (2023) Changes in Erythropoiesis in Type 3 Hemochromatosis Mouse Models. J Mol Pharm Org Process Res 11: 174.

Materials and Method

On the 129X1/svJ strain, two Tfr2 mice models were studied: Tfr2 KI, which has the Tfr2 isoform inactivated, and Tfr2 KO, which has both Tfr2 isoforms inactivated. Tfr2 isoforms were selectively targeted using the same target design in which the murine M163K mutation was introduced in murine Tfr2 gene exon 4 flanked by three loxP sites activated by the Cre/lox recombination system.

Tfr2 KO mice have normal Hamp and serum iron parameters, but splenic iron overload, whereas Tfr2 KI mice have normal Hamp but high serum ferritin and transferrin saturation, as well as hepatic iron overload.

All animals were housed at the University of Torino's Department of Veterinary Sciences. All experimental techniques and animal housing were carried out in compliance with European and national legislation for the protection of animals used for scientific reasons [6].

The animals' peripheral blood was subjected to a full blood cell count examination. An ADVIA®120 Haematology System was used to measure haemoglobin concentration, hematocrit, erythrocyte number, and other indices.

Spleen and bone marrow from killed mice were collected and used for flow cytometric analysis with APC-Ter119 and PE-CD71. Fc-receptors were previously blocked using anti-mouse CD16/CD32 antibodies; leukocytes were excluded using a combination of FITC-conjugated lymphoid and myeloid markers, and dead cells were excluded using 7-AAD. EBiosciences provided all of the reagents.

Ter119-positive events were divided into five subsets, each reflecting a different stage of development, based on CD71 intensity and FSC characteristics. Hematological variables: Blood is drawn from experimental and control mice, and several hematological parameters are examined. Complete blood cell counts, haemoglobin levels, hematocrit, mean corpuscular volume, mean corpuscular haemoglobin, and mean corpuscular haemoglobin concentration are examples of these. These measurements shed light on how TfR2 deficiency affects erythropoiesis and red blood cell properties [7].

To determine iron levels, tissue samples from the liver and spleen are obtained. Iron deposition and iron levels can be assessed using techniques such as Perls' Prussian blue staining or atomic absorption spectrometry.

Using density gradient centrifugation or magnetic cell sorting procedures, bone marrow and spleen samples are taken, and erythroid cells are separated. Flow cytometry can be used to characterise erythroid progenitor populations such as common myeloid progenitors, megakaryocyte-erythroid progenitors, and erythroid precursors at different stages of development.

Using commercially available kits, total RNA is extracted from isolated erythroid cells or tissue samples. Using reverse transcriptase and oligo primers, reverse transcription converts RNA into complementary DNA.

Quantitative Real-Time PCR is used to assess gene expression levels in erythropoiesis, iron metabolism, and inflammatory signaling pathways. SYBR Green or TaqMan probes are employed for detection, and gene-specific primers are created. The levels of gene expression are assessed and normalized to housekeeping genes [8].

Discussion

Iron is well known to be necessary for proper erythropoiesis. The most critical process that is compromised in iron deficiency is RBC formation, which occurs first in the bone marrow and then in the spleen. Erythropoiesis experiences physiological changes that reflect an organism's needs throughout its existence. It increases during adolescence, when there is rapid physical growth, but it remains relatively constant during adulthood and tends to diminish with age. The regulation of hepcidin, one of the main iron regulators, ensures appropriate iron availability for this dynamic erythropoiesis.

Transferrin receptor 2 alpha has been found to function as an iron sensor in the liver as well as an erythropoiesis regulator in erythropoietic tissues. Notably, the gene encoding TFR2 is translated in two major isoforms: the alpha form, which is expressed in the liver and a few other tissues, and the shorter beta form, which is expressed in a limited number of tissues. It is found in substantially higher amounts in the spleen, though. Tfr2 influences the plasma membrane of the liver. It does not respond directly to iron levels, but it is stabilised on the plasma membrane by iron-loaded transferrin. According to the most recent functional models, hepatic Tfr2 interacts with other iron proteins such as Tfr1 and Hfe to sense body iron levels and transduce the signal of iron excess via the Smad 1/5/8 and/or Erk1/2 pathways, resulting in an increase in hepatic hepcidin [9].

As a result, the goal of this manuscript was to investigate the involvement of both Tfr2 isoforms in erythropoiesis as well as the relevance of available iron in erythropoiesis modulation. We employed Tfr2 KI animals with normal circulating iron levels and Tfr2 KO mice with severe iron overload as well as elevated serum ferritin and transferrin saturation. We compared these animals' erythropoiesis to that of their WT littermates. Furthermore, we investigated these two Tfr2 mouse models at young age, when iron demand is high to meet development requirements, and at mature age, when iron is predominantly required for erythropoiesis maintenance.

Our data suggest that adult Tfr2 KO mice had normal erythroid parameters at CBC, with the exception of increased MCV and haemoglobin content. This suggests that in Tfr2 knockout animals, the most HB is created in the RBC during the early phases of erythropoiesis, when cells are bigger. In contrast, the same quantity of haemoglobin reaches the final concentration in WT animals due to a reduction in RBC dimension. This behaviour could be related to the body's desire to eliminate excess iron.

According to earlier research, the left shift in the maturation sequence could be indication of delayed erythropoiesis. The absence of reticulocytosis in these mice, on the other hand, can be explained by an increase in total BM apoptosis, as verified by an increase in the apoptotic marker Caspase-3 and a parallel decrease in the antiapoptotic protein Bcl. This could be a late-stage regulatory mechanism that accounts for the depletion of late precursors, which leads to inefficient erythropoiesis [10].

Conclusion

Type 3 hemochromatosis mouse models have shed light on the complex link between altered erythropoiesis and iron overload. These models have provided vital insights into the underlying mechanics of this illness as well as potential therapy methods. More study is needed to completely understand the complicated biological mechanisms involved and convert these results into effective treatments for type 3 hemochromatosis patients. Tfr2 deficiency in the germline produces adult erythropoiesis in both the BM and the spleen of young mice. In contrast, Trf2 deficiency causes enhanced but immature splenic erythropoiesis, which normalises during animal growth.

Conflicts of Interest

None

Acknowledgment

None

References

1. Liu ZX, Liu SL, Yang HQ (2013). Using protein sequence-structure features. J Comput Theor Nanosci 12: 3023-3027.
2. Li L, Luo Q, Xiao W (2016) A machine-learning approach for predicting palmitoylation sites from integrated sequence-based features. J Bioinform Comput Biol 15: 120-127.
3. Shao-Wu Z, Li-Yang H, Ting-He Z (2014) Prediction of protein-protein interaction with pairwise kernel support vector machine. Int J Mol Sci 15: 3220-3233.
4. Ng SK, Zhuo Z, Soon-Heng T, Kui L (2003) InterDom a database of putative interacting protein domains for validating predicted protein interactions and complexes. Nucleic Acids Res 31: 251-254.
5. Musculoskeletal C, Wenham Y, Conaghan PG (2010) The role of synovitis in osteoarthritis. Ther Adv Dis 2: 349-359.
6. Lajeunesse D, Massicotte F, Pelletier JP, Martel-Pelletier J (2003) Subchondral bone sclerosis in osteoarthritis: not just an innocent bystander. Mod Rheumatol 13: 7-14.
7. Jevsevar DS (2013) Treatment of osteoarthritis of the knee: evidence-based guideline, 2nd edition. J Am Acad Orthop Surg 21: 571-576.
8. Vane JR (1971) Inhibition of prostaglandin synthesis as a mechanism of action for aspirin-like drugs. Nat. Cell Biol 231: 232-235.
9. Scott DL, Berry H, Capell H (2000) The longterm effects of nonsteroidal antiinflammatory drugs in osteoarthritis of the knee: a randomized placebo controlled trial. Rheumatology 39: 1095-1101.
10. Shao-Wu Z, Li-Yang H, Ting-He Z (2014) Prediction of protein-protein interaction with pairwise kernel support vector machine. Int J Mol Sci 15: 3220-3233.