

Renal cells that produce erythropoietin: Their physiology and pathology

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ABSTRACT

Patients with Chronic Kidney Disease (CKD) frequently experience anemia, which raises mortality and morbidity rates. Even though our understanding of the underlying mechanisms of erythropoiesis has significantly improved, Erythropoietin (EPO) stimulating medications are currently the only option for treating renal anemia. This article's goal is to review the physiology of erythropoiesis, the functional significance of EPO, and the underlying molecular and cellular mechanisms that control the synthesis of EPO. At the mRNA level, EPO synthesis is regulated. Hypoxia Inducible Factor (HIF), a transcriptional factor, attaches to the EPO 5' hypoxic response element when anemia or hypoxia takes place, increasing EPO gene transcription.

Pericytes are the principal producers of renal EPO. Renal anemia develops in CKD as a result of pericytes trans differentiation into myofibroblasts and consequent decline in EPO production capacity. Recent experimental and clinical research demonstrates the prolyl hydroxylase inhibitors promise effectiveness in treating renal anemia by enhancing EPO synthesis through stabilizing HIF. The investigation of EPO gene expression at the chromatin level is now possible thanks to recent developments in epigenetics. We'll talk about how restoring EPO expression is impacted by demethylating agents, offering a fresh method for treating renal anemia.

Keywords: Chronic kidney disease; Epigenetics; Erythropoietin; Hypoxia inducible factor; Myofibroblast pericyte

INTRODUCTION

Through the process of hematopoiesis, Hematopoietic Stem Cells (HSCs) are the stem cells that give rise to both the myeloid and lymphoid lineages of blood cells. The fate of HSCs is hypothesized to be determined by the transcription factors PU.1 and GATA1. HSCs develop into megakaryocytic erythroid progenitors with increased expression of GATA1 and decreased expression of PU.1 (MEPs). Megakaryocytic lineage and erythroid lineage need to be further separated, and one of the most crucial components is erythroid Kruppel-Like Factor 1. (KLF1). MEPs develop into the earliest progenitor committed to the erythroid lineage, burst forming unit erythroid, under the direction of KLF1 (BFU-E). Stem Cell Factor (SCF) and insulin like growth factor I are two factors that encourage the proliferation of BFU-E. (IGF1). Colony forming unit erythroids are the erythroid progenitors subsequent stage (CFU-E). Rapidly dividing cells containing the EPO receptor called CFU-E progenitors (EPOR). They grow progressively less susceptible to SCF and IGF1, but very responsive to EPO. Proerythroblasts are produced by CFU-E and develop into basophilic, polychromatic, and then orthochromatophilic erythroblasts in that order. Orthochromatophilic erythroblasts are enucleated, and in the bone marrow, they eventually develop into reticulocytes. Reticulocytes are then released into circulation and purge all extra organelles to create erythrocytes [1].

LITERATURE REVIEW

CFU-E, proerythroblasts, and early basophilic erythroblasts all express EPOR, a transmembrane glycoprotein. These erythroid progenitors are shielded from apoptosis by EPO binding. Since the stage of late basophilic erythroblasts, the amount of EPOR has continuously decreased, and it is absent on more mature forms of erythroblasts. As soon as EPO binds to EPOR, the protein undergoes a conformational shift that causes Janus kinase 2 to become activated and phosphorylate tyrosine in the cytoplasmic domain of EPOR. This then activates the downstream signals, including Ras, phosphoinositol-3 kinase/AKT kinase, and Signal transducer and activator of Transcription 5 (SIRT5).

EPO's binding to EPOR also causes the endocytosis of EPO-EPOR complexes, which are ultimately broken down in lysosomes and use the body's supply of EPO. When a patient recovers from an acute blood loss, this process provides a negative feedback to reduce EPO level and prevent an excess of erythrocyte formation [2].

Regulation of EPO gene

In a fetus, the liver produces EPO. After birth, the kidney takes over as the primary source of EPO production. At the mRNA level, the kidney's ability to produce EPO is controlled. In rat investigations, the degree of anemia was correlated with the expression of Epo mRNA, which increased following phlebotomy. Additionally, cobalt administration to rodents increased the expression of renal EPO mRNA. According to this research, EPO transcription can be affected by hypoxic stress and anemia, and there may be a cellular oxygen sensing system that regulates EPO transcription [3].

In order to determine the regulation of EPO transcription, Semenza and colleagues created different lines of transgenic mice. Transgenic mice with a 4 kb DNA fragment (EPO4Tg) containing the human EPO gene, 0.4 kb 5' flanking sequence, and 0.7 kb 3' flanking sequence developed polycythemia due to the widespread expression of the EPO gene in unsuitable tissues. Notably, anemia in these animals can generate EPO mRNA in the liver but not the kidney [4].

Transgenic animals carrying the human EPO gene, 6 kb 5' flanking sequence, and 0.7 kb 3' flanking sequence (EPO10Tg) were created by extending this construct with an additional 6 kb 5' flanking region. These polycythemic mice preserved the ability to induce the liver but had their ectopic EPO production in other tissues reduced. This suggests that a negative regulatory region between 0.4 kb and 6 kb in the 5' flanking sequence was responsible for this [5].

of cardiovascular conditions were extracted and compared in order to these transgenic mice were able to be stimulated EPO mRNA expression by anemia and hypoxia both in the kidney and liver when this DNA construct was further extended to an 18 kb human EPO gene (EPO18Tg) by

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additional 14 kb 5' flanking region. Compared to EPO4Tg and EPO10Tg mice, EPO18Tg mice displayed a higher level of polycythemia because kidneys are the primary location of adult EPO production [6].

These findings showed that the DNA elements controlling kidney and liver EPO synthesis are located in separate places. The kidney inducible element is situated between 9.5 kb and 14 kb 5' flanking sequence, while the liver inducible element is found within 0.7 kb 3' flanking sequence [7].

It was discovered that two human hepatoma cell lines, hep3B and hepG2, produced EPO when exposed to hypoxia, furthering the *in vitro* investigation of the Hypoxic Response Element (HRE) in liver cells [8].

Then, Hypoxia Inducible Factor (HIF), a transcription factor, was discovered. HIF was made to attach to the 3' end of the EPO gene as a result of hypoxia, which activated the gene's transcription. The minimal sequence needed for HIF dependent transcriptional activation was found at the 5'-RCGTG-3' motif on the 3' end of the EPO gene, according to further research of HIF binding motifs. Nine base pairs downstream of the HIF binding site, a CACA repeat sequence was also discovered. The CACA repeat is an accessory sequence required for the full functionality of oxygen related EPO transcription, as evidenced by the fact that a mutation of this sequence abolished the induction of EPO by hypoxia in hepatoma cell line [9].

Hypoxia inducible factor

Basic helix loop helix heterodimer protein HIF is a member of the PAS (PER-ARNT-SIM) family. Numerous developmental and physiological processes, including erythropoiesis as well as energy metabolism, angiogenesis, iron homeostasis, cell proliferation, and differentiation, are influenced by HIF binding across the genome. A subunit (HIF) and a subunit (HIF), also known as the aryl hydrocarbon nuclear translocator, make up the HIF complex (ARNT). HIF is constitutive, whereas HIF is strongly induced by hypoxia and sensitive to oxygen. HIF1, HIF2, and HIF3, three distinct HIF proteins that dimerize with HIF, have all been discovered. More and more research on animals has shown that HIF2 regulates EPO transcription more effectively than HIF1 does. Mice with postnatal HIF2 ablation developed anemia [10].

Oxygen sensing mechanism

The secret to oxygen sensing is determined by HIF's rapid breakdown. The HIF subunit rapidly degrades in normoxia but is stable under hypoxia. This mechanism has been further clarified as a result of the discovery of the Von Hippel-Lindau (VHL) protein. When VHL was absent from cells, HIF was expressed by default. Under normoxia, the proline residue of the HIF subunit is hydroxylated. The VHL/E3 ubiquitin ligase complex can identify and polyubiquitinate this hydroxylated HIF, which the proteasome can then destroy.

Once hypoxia is present, proline hydroxylation is suppressed, inhibiting the breakdown of HIF. Stabilized HIF translocates into the nucleus, forms a heterodimer with HIF, and subsequently activates target genes transcriptionally.

Renal erythropoietin producing cell

Renal tubular epithelial cells were once mistakenly identified in various investigations as Renal EPO Producing Cells (REPCs). REPCs are found between tubular cells of the outer medulla and cortex, namely the interstitium, according to research using *in situ* hybridization of EPO mRNA. Research using transgenic mice showed that REPCs are interstitial fibroblast like cells and are polymorphic in shape with linking cytoplasmic processes.

Mechanisms of renal anemia in chronic kidney disease

Anemia in CKD is influenced by a number of factors. Renal anemia was thought to be caused by circulating uremia related inhibitors of erythropoiesis, but recombinant human EPO's ability to treat renal anemia

proved that this mechanism was rather unimportant. EPO deficiency is the most significant of the factors that contribute to renal anemia. Reduced oxygen sensing and EPO synthesis in REPCs may be the cause of EPO deficiency.

Regardless of the underlying condition that initially causes kidney injury, interstitial fibrosis is present in all cases of CKD. As a result, normal kidney tissue and function are irreversibly lost. As previously mentioned, in CKD, the REPCs, also known as fibroblasts or pericytes, developed into myofibroblasts.

DISCUSSION

Treatment of renal anemia by oxygen sensing mechanism

Erythropoiesis Stimulating Agents (ESAs) and iron supplements are being used to treat renal anemia. The development of a novel therapeutic strategy for renal anemia, however, represents an unmet clinical need due to various clinical concerns with the use of exogenous ESA, such as the increased risk of death and cardiovascular events in patients with poor response or malignancy. The PHD inhibitors offer a fresh possibility for treating renal anemia. PHD inhibitors boost the transcription of EPO mRNA in REPCs *via* activating the HIF pathway by blocking the proteasomal degradation of HIF.

CONCLUSION

In the past, ESA and iron supplements were necessary for the treatment of renal anemia in CKD. By improving oxygen sensing mechanisms and altering epigenetic patterns, recent developments in our understanding of the physiology and pathophysiology of REPCs open up possibilities for the development of novel therapeutic approaches. The recently created PHD inhibitors improve the oxygen sensing mechanism by stabilizing HIF, which therefore promotes the generation of EPO. The 5' regulatory region of the EPO gene undergoes hypermethylation during kidney fibrosis as pericytes transition to myofibroblasts, limiting HIF binding and subsequently EPO transcription. The creation of demethylating agents or epigenetic editing that targets the EPO 5' regulatory element of REPCs may represent a future therapeutic strategy.

REFERENCES

1. Chou YH, Huang TM, Chu TS, et al. Novel insights into acute kidney injury chronic kidney disease continuum and the role of renin angiotensin system. *J Formos Med Assoc.* 2017;116(9):652-9.
2. Xia H, Ebben J, Ma JZ, et al. Hematocrit levels and hospitalization risks in hemodialysis patients. *J Am Soc Nephrol.* 1999;10(6):1309-16.
3. Chou YH, Tsai TJ. Autonomic dysfunction in chronic kidney disease: An old problem in a new era. *J Formos Med Assoc.* 2016;115(9):687-8.
4. Romijn JA. Erythropoietin. *N Engl J Med.* 1991;325: 1176-7
5. Burda P, Laslo P, Stopka T, et al. The role of PU. 1 and GATA-1 transcription factors during normal and leukemogenic hematopoiesis. *Leukemia.* 2010;24(7):1249-57.
6. Koury MJ, Bondurant MC. Erythropoietin retards DNA breakdown and prevents programmed death in erythroid progenitor cells. *Science.* 1990;248(4953):378-81.
7. Gross AW, Lodish HF. Cellular trafficking and degradation of erythropoietin and Novel Erythropoiesis Stimulating Protein (NESP). *J Biol Chem.* 2006;27;281(4):2024-32.
8. Wenger RH, Stiehl DP, Camenisch G, et al. Integration of oxygen signaling at the consensus HRE. *Sci STKE.* 2005 (2005): 12

9. Wang GL, Semenza GL. General involvement of hypoxia inducible factor 1 in transcriptional response to hypoxia. Proc Natl Acad Sci USA. 1993;90(9):4304-8.
10. Yi X, Liang Y, Huerta-Sanchez E, et al. Sequencing of 50 human exomes reveals adaptation to high altitude. Science. 2010;329(5987):75-8.