eISSN (Online): 2598-0580

Bioscientia Medicina: Journal of Biomedicine & Translational Research

Journal Homepage: www.bioscmed.com

Control of HIF-1α Levels Potentially Promotes the Tissue Repair in Various Conditions Through Target Gene Expression

Riyadh Firdaus[1], Ani Retno Prijanti[2*]

[1] Department of Anesthesiology and Intensive Care, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia
[2] Department of Biochemistry and Molecular Biology, Faculty of Medicine, Universitas Indonesia, Jakarta, Indonesia

ARTICLE INFO

Keywords:
HIF-1α
Hypoxia
Gene expression
Tissue repair
Cancer

***Corresponding author:**
Ani Retno Prijanti

E-mail address:
aniretno@gmail.com

All authors have reviewed and approved the final version of the manuscript.

https://doi.org/10.32539/bsm.v6i1.429

ABSTRACT

Hypoxia inducible factor-1 (HIF-1) is a transcription factor that plays an important role in maintaining oxygen balance at both the cellular and systemic levels, and is associated with various controls in the body. HIF-1 is a heterodimer of alpha and beta subunits. Alpha subunits are mostly dependent on oxygen levels in the body. In many cancers, excessive HIF-1α is thought to be involved in the promotion of tumor growth and metastasis. In addition, in the induction of systemic hypoxia, there is an increase of HIF-1α in the heart, brain, and even the kidneys as an adaptation response to hypoxia. Several studies regarding HIF-1a expression in traumatic brain injury, found that HIF-1a increased immediately after TBI, and decreased significantly after 24 hours. This can be used as a basis for further research on HIF-1a control as an effort to stop tissue damage or even help tissue repair.

1. Introduction

Hypoxia is a state of low oxygen concentration in cells or tissues that can threaten the cell survival. Aerobic organisms, from prokaryotes to eukaryotes, possess adaptive homeostatic mechanisms to overcome hypoxia at both the cellular and systemic levels through oxygen sensing. At the cellular level, decreased oxygen levels result in the activation of several metabolic pathways that do not require oxygen (induction of the anaerobic glycolysis enzyme). At the systemic level, regulation is performed with the aim of increasing oxygen distribution, such as induction of erythropoiesis, angiogenesis, and hyperventilation.[1]

Hypoxia inducible factor-1 (HIF-1) is transcription factor that has important role in maintaining oxygen balance at the cellular and systemic levels. The genes regulated by HIF-1 are related to vasomotor regulation, angiogenesis, red blood cell formation and iron metabolism, cell proliferation, and energy metabolism.[1,2] Due to so many genes become the targets of HIF-1 and most are the genes that are vital in body function, so this transcription factor has an important role, especially in hypoxic conditions.[3]

The HIF-1 molecule is a heterodimer consisting of α-subunit and β-subunit known as ARNT (Aryl Hydrocarbon Nuclear Translocator). The subunit (HIF-1β) is presented in the nucleus and is constitutively

expressed. Therefore, its activity is not affected by hypoxic conditions. Meanwhile, the -subunit (HIF-1α) which is specifically induced in an adaptive response to hypoxia is believed to be the main regulator of oxygen homeostasis.[2] The stability and activity of the α-subunit is strongly influenced by oxygen level and is regulated by several post-translational modifications. Under normoxia conditions, the α-subunit will undergo hydroxylation at specific proline residues by HIF-prolyl hydroxylase, which causes ubiquitination so that the α-subunit is degraded by proteasomes. On the other hand, under hypoxic conditions, inhibition of hydroxylation of proline residues will stabilize HIF-1α, thereby increasing its activity as a transcription factor for genes that regulated by hypoxia. Thus, the activity of HIF-1 in oxygen homeostasis is largely determined by the α-subunit which being maintained in a hypoxic state and degraded in normoxia state. In addition, a study by Jing et al reported that there was an increase in the expression of HIF-1α mRNA in acute hypoxic conditions.[4]

Hypoxia has been shown to have an important stage in the pathogenesis of various degenerative diseases, especially those caused by ischemia, including myocardial infarction, stroke, cancer, chronic obstructive pulmonary disease, and acute kidney failure.[5] Therefore, information regarding the expression of HIF-1α as a response of cell adaptation in hypoxic conditions is needed to improve the management process of these diseases.

Hypoxia inducible factor-1α (HIF-1α)

Hypoxia can be detected by a marker of hypoxic tissue, namely Hypoxia Inducible Factor 1 or HIF-1 which is a transcription factor that plays a role in maintaining oxygen balance, both cellular and systemic levels. The HIF-1 molecule is a heterodimer consisting of an α-subunit and an β-subunit. There are three HIF-α isoforms (HIF-1α, HIF-2α and HIF-3α). The beta class includes HIF-1β. HIF-1 is a combination of HIF-1α (120 kDa) and HIF-1β (91-94 kDa) subunits. The HIF-1β subunit is a constitutively expressed protein, but the expression of the HIF-1α subunit (cytosol protein) is largely dependent on oxygen levels. Thus, this subunit is believed to be a marker of tissue that is undergoing hypoxia.[3]

Hypoxia Inducible Factor-1 Alpha, also known as HIF-1α, is a subunit of the heterodimeric transcription factor HIF-1 and is considered as the master of transcriptional regulator of cellular responses and development of hypoxia.[1,6] In hypoxic conditions, there is dysregulation and an excessive increase in HIF-1 levels which are widely involved in the biological process of cancer and a number of other pathophysiological processes, especially in vasomotor regulation (NOS2), angiogenesis (VEGF, FLT-1), red blood cell formation and iron metabolism (EPO, transferrin receptor, ceruloplasmin), cell proliferation (IGF-1, IGFBP-1, TGF β), and energy metabolism (GLUT 1-3, phosphoenolpyruvate carboxykinase, lactate dehydrogenase A, aldolase, phosphoglucokinase 1, pyruvate kinase, enolase , prolyl 4-hydroxylase, and adrenomedullin).[4,7] Because there are so many genes that become the target of HIF-1 and most of them are the genes that are vital in body function, this transcription factor has an important role, especially in hypoxic conditions.[4,8]

Hypoxic response pathway

Gregg L, et al (2016) found a pathway involving HIF that plays an important and central role in the oxygen-sensing process of eukaryotic cells.[9] Changes in oxygen availability conditions are an important physiological stimulus for all multicellular organisms, which must match oxygen supply with cell respiration needs. Apart from contributing to the maintenance of intracellular bioenergy by producing mitochondrial ATP, oxygen also functions as a universal electron acceptor in various biochemical pathways. Therefore, the response of cells to hypoxic conditions is very rapid. Hyperoxia is also a condition that should be avoided because it can cause oxidative damage to lipids, proteins, and other biomolecules. Acute responses usually involve changes in the activity of pre-existing molecules, such as ion channels, whereas long-term adaptation involves larger changes, including changes in the programming of global gene expression.[10]

Two important studies described by Ratcliffe and Kaelin (2001) showed that the interaction between the von Hipper-Lindau/pVHL protein and the HIF-1α

subunit depends on the oxygen-mediated hydroxylation of the two proline groups of each HIF-1α subunit. Through genetic approach research with samples of the roundworm *Caenorhabditis elegans*, Ratcliffe's team found that the mutant of VHL-1 always expresses HIF-1α, just like worms that have a deficiency in the gene encoding the dioxygenase enzyme. They found that this enzyme hydroxylated proline residues in the HIF-1α subunit, leading to the instability of HIF-1α when oxygen levels were sufficiently abundant, and also that the action of this enzyme was rapidly inhibited when oxygen levels decreased. The amino acid sequence of the worm dioxygenase enzyme allowed the identification of its three mammalian homologues, namely the prolyl-hydroxylase domain/PHD protein.[11]

A large study led by Semenza, Ratcliffe, and Kaelin was able to elucidate the mechanism of oxygen depletion resulting in decreased proline HIF-hydroxylation process due to PHD inhibition, reversible stabilization of HIF-1α, and stimulation of 500-1000 oxygen-regulated genes which in turn can mediate various adaptive response to hypoxic conditions at the cellular, tissue, and organismal levels.[11] HIF-1α is rapidly upregulated, stabilized, and travels to the nucleus in response to hypoxia. In contrast, HIF-1α degraded rapidly on reoxygenation/reperfusion and under normoxia (Figure 1). Under normoxia, HIF-1α is bound by pVHL. pVHL recruits ubiquitin ligase that targets HIF-1α for 26S proteasomal degradation. The binding of pVHL depends on the hydroxylation of specific proline residues in HIF-1α (pro402 and pro564) by the PHD family of proteins (PHD). PHD uses oxygen as a substrate; therefore, its activity is inhibited under hypoxic conditions. Oxygen can also activate FIH (Factor Inhibiting HIF), preventing the binding of co-activator p300/CBP, thereby decreasing HIF-1-induced transcriptional activity. Reactive oxygen species such as peroxynitrite can damage HIF-1α by oxidizing its thiol group, which is further degraded by the 20S proteasome system.[12]

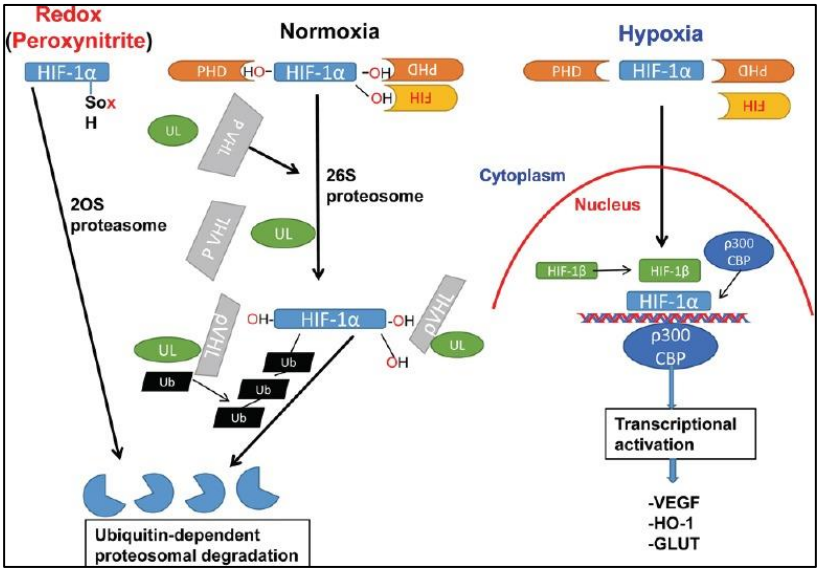

Figure 1. Hypothesis Regulation of HIF-1α in hypoxic, normoxia, and redox states.[12]

Although many other intracellular processes (such as ion channel activity, signal transduction, and oxygen-dependent biochemical reactions) can be sensors of oxygen availability, the HIF-PHD-pVHL axis has a central role with a wide range of linking external oxygen concentration stimuli to nuclear gene expression. The extensive transcriptional output of HIF is highly integrated with additional signaling pathways, nutrient sensing, protein synthesis, and other important processes that depend on cellular ATP levels. The clinical relevance of this oxygen-sensing system is substantial, that is, hypoxic conditions influence many disease progression processes, such as cancer, heart disease, pulmonary hypertension, stroke, bacterial

infection, inflammation, and wound healing.[3,13]

HIF-1α in tumors

HIF-1α is expressed in many types of cancer in humans. HIF-1α overexpression is highly involved in the promotion of tumor growth and metastasis through its role in initiating angiogenesis and regulating cell metabolism to overcome hypoxia. Hypoxia promotes apoptosis in normal cells and tumor cells. However, hypoxic conditions in microtumours together with accumulation of genetic alternation often lead to overexpression of HIF-1α.[14,15]

Significantly elevated levels of HIF-1α were found in solid tumors such as the colon, breast, pancreas, kidney, prostate, ovary, brain, and bladder. Clinically, elevated levels of HIF-1α in a number of cancers, including cervical cancer, non-small cell lung cancer, breast cancer, oligodendroglioma, oropharyngeal cancer, ovarian cancer, endometrial cancer, esophageal cancer, neck cancer, and gastric cancer, has been associated with aggressive tumor progression, and is implicated as a predictive and prognostic marker for resistance to radiotherapy, chemotherapy, and increased mortality.[9,15]

HIF-1α expression can also regulate breast tumor development. High levels of HIF-1α can be detected in early cancer progression and have been found in early ductal carcinoma in situ and pre-invasive stages in breast cancer progression.[16] The occurrence of hypoxia in tumor cells may be due to the presence of necrotic areas, blocking of blood vessels by tumor cells from circulation, collapse of blood vessels due to interstitial pressure, or interruption of tumor blood flow. A study by Hardiany et al found that the relative levels of HIF-1α mRNA were higher in high-grade gliomas of malignancy. In addition, high levels of HIF-1α in glioma cells can also reduce the sensitivity of glioma cells to radiotherapy because they require oxygen.[17]

Bachtiary, et al reported that in cervical cancer, HIF-1α overexpression was significantly associated with radiation response in which patients with HIF-1α overexpression gave only a partial response to radiation therapy. In addition, elevated levels of HIF-1 in cervical cancer and oropharyngeal cancer were associated with patient survival, whereas patients who expressed higher HIF-1α had lower survival rate.[15]

Studies of glioblastoma multiforme have shown striking results between HIF-1α expression patterns and VEGF gene transcription. This further demonstrates the regulatory role of HIF-1α in promoting tumor development, possibly via a hypoxia-induced VEGF expression pathway.[9] HIF-1α overexpression in tumors may also occur in an independent hypoxia pathway. In hemangioblastoma, HIF-1α expression was found in most well-vascularized tumor sample cells. In both renal carcinoma and hemangioblastoma, the von Hippel-Lindau gene is inactivated, and HIF-1α is expressed at high levels. In addition to VEGF overexpression in response to elevated HIF-1α levels, the phosphatidylinositol 3-kinase/PI3K-AKT pathway is also involved in tumor growth. Phosphatase and tensin homologous deleted on chromosome 10 (PTEN) mutations are common in prostate cancer and are associated with tumor progression to an aggressive stage, increased vascular density, and angiogenesis.[18]

Expression of HIF-1α towards vascular endothelial growth factor (VEGF)

From a study about the expression of HIF-1α in astrocytoma, it stated that the formation of new blood vessels (angiogenesis) was found. The process of angiogenesis occurs as a result of an imbalance of pro- and anti-angiogenic factors. One of the pro-angiogenic factors is Vascular Endothelial Growth Factor (VEGF) which is produced by cancer cells, stromal tissue, and macrophages due to chronic inflammatory processes in tumors. VEGF which is also known as Vascular Permeability Factor (VGF) is important in the angiogenesis process that underlies tumor growth and the formation of peritumoral edema in brain tumors. Angiogenesis is a process of pro- and anti-angiogenic balance of tumor cells and normal cells. The binding between VEGF and VEGFR-1 and 2 will cause the process of angiogenesis. VEGF expression in both benign and malignant tumor cells is highly variable.[14,19]

One of the causes of angiogenesis induction in tumor growth is the rapid tumor proliferation, which can eventually lead to a hypoxic state due to the limited

oxygen diffusion due to tissue growth. To get an adequate supply of nutrients and oxygen in tumors larger than 2-3 mm^3, new blood vessels are needed. Hypoxia is an important stimulus for tumor angiogenesis resulting in the stabilization of HIF-1α. HIF-1α expression will increase and form dimerization with HIF-1β, then this complex binds to DNA, recruits co-activators and transcripts the target genes. HIF-1 target genes will encode pro-angiogenic growth factors such as VEGF.[20]

The transcription factor NF-kB which can be activated in hypoxic conditions through the formation of ROS in mitochondria can regulate VEGF expression, while the Ets transcription factor can regulate various target genes downstream in endothelial cells that promote the formation of angiogenic phenotype, including the increase in VEGF-R, urokinase, and various matrix metalloproteinases (MMPs). VEGF binds to VEGFR-1 and VEGFR-2 for the process of angiogenesis. This binding activates signaling pathways resulting in the increase of gene expression that involved in the proliferation and migration of endothelial cells, promoting cell survival, and vascular permeability. VEGF also has a considerable influence on tumor growth and the formation of peritumoral edema. Several previous studies stated that high grade astrocytoma had a higher angiogenesis rate than low grade astrocytoma. VEGF expression has an influence on various levels of malignancy in neuroblastoma, prostate and lung cancer. The study of Jakovljevic et al stated that the results showed that high VEGF expression was associated with an unfavorable histological type of neuroblastoma. In prostate adenocarcinoma, VEGF expression has an effect on increasing Gleason scores.[20]

The expression of VEGF in lung tumors type non-small cell carcinoma also increased significantly according to the tumor size and the tumor cell proliferation index. Previous studies on cerebral edema have shown that the expression of VEGF and Aquaporin 4 is associated with peritumoral edema in meningiomas. VEGF expression has a role in increasing vascular permeability while Aquaporin 4 acts as a transmembrane fluid transport. The effect of VEGF is still a matter of debate because other studies have shown that some gliomas with extensive peritumoral edema show low VEGF expression. Until now, there has been no study regarding the relationship between VEGF expression level with histopathological grade, tumor size, and incidence of peritumoral edema in astrocytoma.[21]

Relative expression of HIF-1α mRNA in mice heart, brain, blood and kidneys during induction of systemic hypoxia

Induction of systemic hypoxia caused a gradual decrease in the pO_2 and pCO_2 of the rat blood, as well as a decrease in the blood hematocrit which indicated a decrease in the number of red blood cells in the blood. On the Melting Curve (Figure 2), only one peak was seen for HIF-1α (79.5°C) and Beta-actin (85.5°C). This shows that the primer pairs used for the two genes are quite good, specific, and do not cause primary dimers. In addition, the DNA electrophoresis image also shows only one DNA band (HIF-1α: 174 bp; Beta-actin: 174 bp) which proves that the cDNA amplification product is matching as targeted by primer pair of each gene.[22,23]

In Figure 2 it appears that the relative mRNA levels of HIF-1α in the heart and brain of rats induced by systemic hypoxia gradually increased. The increase reached its peak on day 7 in the heart and day 14 in the brain. After that, it began to decrease slightly. This suggests that the responsive pathway for adaptation to early hypoxia is acquired through increased expression of the HIF-1α gene. In further hypoxia, it seems that the increased expression of the HIF-1α gene is slowly replaced by the stabilization of the HIF-1α protein, that is, by inhibiting its degradation.

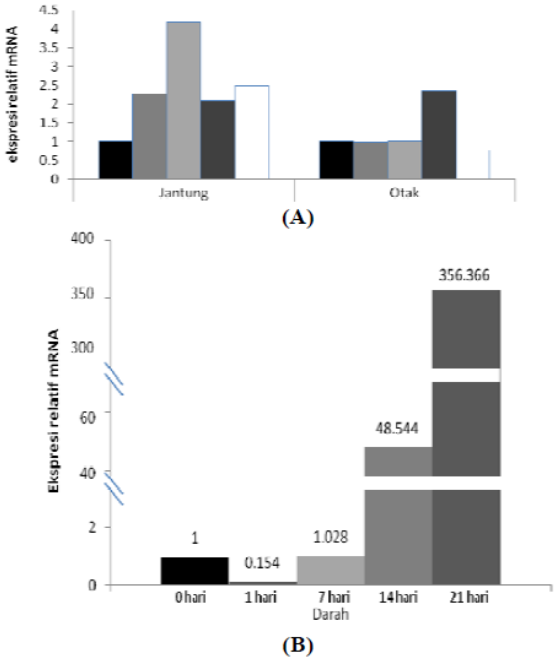

Figure 2. Effect of hypoxia on relative expression of HIF-1α mRNA in heart and brain (A), and blood (B). Relative expression of HIF-1α mRNA in mice after 1, 7, 14 and 21 days of hypoxia normalized by expression in the group without hypoxia (0 days).[22]

Shao, et al also reported that HIF-1α mRNA expression was increasing in acute hypoxia, while in repeated hypoxia it decreased again. Although there was an increase of HIF-1α protein in repeated hypoxia due to inhibition of degradation, however, the results of this study showed that the stimulation of HIF-1α expression in leukocyte cells remained intensively maintained until day 21 even though its expression had reached very high levels. This is presumably to intensify O_2 uptake and distribution, leukocytes require more adaptive responses through increased of HIF-1α gene expression. Meanwhile, the possibility of inhibition of proteasome degradation for the stabilization of HIF-1α protein is slower than heart and brain. Thus, it can be assumed that leukocytes are slower to adapt to induction of systemic hypoxia. In addition, from the results of this study, it can be observed that the maximum increase of HIF-1α mRNA relative expression in brain tissue was significantly lower than the relative expression of HIF-1α that found in heart and blood.

It indicates that brain tissue is more resistant to systemic hypoxia than heart and blood or it is possible that brain tissue has other adaptive responses to hypoxia other than HIF-1α expression. Analysis of the relationship between changes in the relative expression of HIF-1α mRNA in the heart, brain and blood showed that there was no relationship between blood, heart, and brain. This analysis strengthens the notion that heart, brain, and blood tissues have adaptive responses to hypoxia with different capacity and sensitivity, so that the measurement of the relative levels of HIF-1α mRNA in the blood cannot describe the hypoxic state of the heart and brain.[24]

In a study by Prijanti et al, it was found that the increase of HIF-1α in systemic hypoxia has a strong correlation with renin expression from renal. Although the accumulation of HIF-1α does not directly activate the transcription of renin.[25] It has been known previously that renin secretion can be stimulated by chloride ions in the renal tubules which detected by juxtaglomerular cells (JGCs), but renin secretion can also be stimulated directly by sympathetic nerves to the kidneys via beta-adrenergic receptors on JGCs, where beta-adrenergic receptors mediate hypoxia sensing and this leads to accumulation of HIF-1α.[26]

HIF-1α and traumatic brain injury (TBI)

Immediately after TBI, direct trauma and lack of blood flow lead to necrotic nerve death. However, greater loss of apoptotic neurons may occur later from secondary injury caused by hypoxia/ischemia that associated with oxidative stress and inflammation.[27] Unlike stroke, the role of HIF-1α in TBI is poorly understood. While HIF-1α activity increased immediately after TBI, its expression level decreased significantly 24 hours after TBI.[28] Studies from other researches have also reported that neuroreparative (stimulation of VEGF and brain-derived neurotrophic factor/BDNF) expression in the chronic phase of TBI is dependent on HIF-1α activity.[29] The HIF-1α pathway is involved in pathological (hypoxia) and neuroreparative (normoxia) mechanisms after TBI. HIF-1α stabilizers/inducers, such as desferrioxamine (an iron chelator approved for the treatment of hemochromatosis), promote a number of survival pathways, including neuroprotection, angiogenesis and neurotrophins, and reduce cerebral infarction when administered before or after stroke.[30]

PHD inhibitors, such as FG-4539, are currently in phase II trials due to their activity to stabilize HIF-1α by preventing degradation by the ubiquitin proteasome system.[12] However, inhibition of HIF-1α in the acute injury phase of TBI has also been reported to be neuroprotective. The TBI study by Khan, et al demonstrated that the HIF-1α/VEGF pathway accelerates functional recovery in mice of post 2-week TBI via S-nitrosylation of HIF-1α, which further, supports the neuroreparative role of HIF-1α. GSNO-mediated S-nitrosylation (S-nitrosoglutathione) occurs in pVHL, PHD, and HIF-1α, contributing to the inhibition of HIF-1α degradation. S-nitrosylated HIF-1α travels to the nucleus, where it then dimerizes with HIF-1β and interacts with P300 and CBP, leading to the transcription of neuroreparative mediators such as VEGF and BDNF under TBI conditions.[24,31]

Administration with GSNO accelerates functional recovery and improves overall outcomes in relatively long-term TBI studies. Furthermore, administration of GSNO in humans for other indications did not produce any toxicity or side effects, thus supporting the translational potential of GSNO therapy in TBI. However, long-term studies are needed to evaluate neuroreparative mechanisms and improve neurological function to determine the effectiveness of GSNO.[31]

2. Conclusion

Hypoxia is a state of low oxygen concentration in cells or tissues that can threaten cell survival. HIF-1 is a transcription factor that plays an important role in maintaining oxygen balance at both cellular and systemic levels, and is a master of regulators of genes that are vital in body function. HIF-1α is rapidly regulated, stabilized, and travels to the nucleus in response to hypoxia. In contrast, HIF-1α degraded rapidly on reoxygenation/reperfusion and under normoxic conditions. Although many intracellular processes can be sensors of oxygen availability, the HIF-PHD-pVHL axis has a broad, central role to link external oxygen concentration stimuli to global expression of nuclear genes.

HIF-1α is expressed in various types of cancer in humans. HIF-1α excess may be involved in promoting tumor growth and metastasis through its role in initiating angiogenesis and regulating cell metabolism to overcome hypoxia. It is known that brain tissue is more resistant to systemic hypoxia than other organs. In addition, the discovery of various compounds that can stabilize HIF-1α has potential as a therapy of the disease.

3. Acknowledgment

The authors would express their gratitude to the Doctoral Program in Biomedical Science, Faculty of Medicine, Universitas Indonesia.

4. References

1. Simon MC. The hypoxia response pathways — Hats Off! N Engl J Med. 2016; 375(17): 1687-89.
2. Semenza GL. HIF-1: Mediator of physiological and pathophysiological responses to hypoxia. Journal of Applied Physiology. 2000; 88(4) :1474-80.
3. Semenza GL. HIF-1 and mechanisms of hypoxia sensing. Current Opinion in Cell

Biology. 2001; 13(2): 167-71.
4. Jing X, Yang F, Shao C, Wei K, Xie M, Shen H, et al. Role of hypoxia in cancer therapy by regulating the tumor microenvironment. Molecular Cancer. 2019; 18: 157.
5. Chen PS, Chiu WT, Hsu PL, Lin SC, Peng IC, Wang CY, et al. Pathophysiological implications of hypoxia in human diseases. Journal of Biomedical Science. 2020; 27: 63.
6. Ke Q, Costa M. Hypoxia-inducible factor-1 (HIF-1). Molecular pharmacology. 2006; 70(5): 1469-80.
7. Sen T, Sen N. Treatment with an activator of hypoxia-inducible factor 1, DMOG provides neuroprotection after traumatic brain injury. Neuropharmacology. 2016; 107: 79-88.
8. Liu Y, Cox SR, Morita T, Kourembanas S. Hypoxia regulates vascular endothelial growth factor gene expression in endothelial cells: Identification of a 5' enhancer. Circ Res. 1995; 77(3): 638-43.
9. Semenza GL. The hypoxic tumor microenvironment: A driving force for breast cancer progression. Biochimica et Biophysica Acta - Molecular Cell Research. 2016; 1863(3): 382-91.
10. Semenza GL. Hypoxia-inducible factor 1: Regulator of mitochondrial metabolism and mediator of ischemic preconditioning. Biochimica et Biophysica Acta - Molecular Cell Research. 2011; 1813(7): 1263-68.
11. Kaelin WG, Ratcliffe PJ. Oxygen sensing by metazoans: The central role of the HIF hydroxylase pathway. Molecular Cell. 2008; 30(4): 393-402.
12. Harten SK, Ashcroft M, Maxwell PH. Prolyl hydroxylase domain inhibitors: A route to HIF activation and neuroprotection. Antioxidants and Redox Signaling. 2010; 12(4): 459-80.
13. Marxsen JH, Stengel P, Doege K, Heikkinen P, Jokilehto T, Wagner T, et al. Hypoxia-inducible factor-1 (HIF-1) promotes its degradation by induction of HIF-α-prolyl-4-hydroxylases. Biochem J. 2004; 381: 761-7.
14. Alalem M. The Role of $_{M}$TOR in SAF-1-Mediated VEGF expression and breast cancer progression. ProQuest Dissertations and Theses. 2015.
15. Bachtiary B, Schindl M, Pötter R, Dreier B, Knocke TH, Hainfellner JA, et al. Overexpression of hypoxia-inducible factor 1α indicates diminished response to radiotherapy and unfavorable prognosis in patients receiving radical radiotherapy for cervical cancer. Clin Cancer Res. 2003; 9(6): 2234-40.
16. Liu Z ji, Semenza GL, Zhang H feng. Hypoxia-inducible factor 1 and breast cancer metastasis. Journal of Zhejiang University: Science B. 2015; 16(1): 32-43.
17. Hardiany NS, Sadikin M, Wanandi SI. Ekspresi relatif mrna hypoxia inducible factor-1α pada sel glioma penderita. Indones J Cancer. 2013; 7(1): 1-5.
18. Kimbro KS, Simons JW. Hypoxia-inducible factor-1 in human breast and prostate cancer. Endocrine-Related Cancer. 2006; 13(3): 739-49.
19. Guo H, Zhou H, Lu J, Qu Y, Yu D, Tong Y. Vascular endothelial growth factor: An attractive target in the treatment of hypoxic/ischemic brain injury. Neural Regen Res. 2016; 11(1): 174-9.
20. Palazon A, Tyrakis PA, Macias D, Veliça P, Rundqvist H, Fitzpatrick S, et al. An HIF-1α/VEGF-A Axis in Cytotoxic T Cells Regulates Tumor Progression. Cancer Cell. 2017; 32(5): 669-83.
21. Lin F, Pan LH, Ruan L, Qian W, Liang R, Ge WY, et al. Differential expression of HIF-1α, AQP-1, and VEGF under acute hypoxic conditions in the non-ventilated lung of a one-lung ventilation rat model. Life Sci. 2015; 124: 50-5.
22. Graham AM, Presnell JS. Hypoxia Inducible Factor (HIF) transcription factor family

expansion, diversification, divergence and selection in eukaryotes. PLoS One. 2017; 12(6): e0179545.

23. Khatua S, Peterson KM, Brown KM, Lawlor C, Santi MR, LaFleur B, et al. Overexpression of the EGFR/FKBP12/HIF-2α pathway identified in childhood astrocytomas by angiogenesis gene profiling. Cancer Res. 2003; 63(8): 1865-70.
24. Khan M, Khan H, Singh I, Singh AK. Hypoxia inducible factor-1 alpha stabilization for regenerative therapy in traumatic brain injury. Neural Regeneration Research. 2017; 12(5): 696-701.
25. Prijanti AR, Ranasasmita R, Sandra Y, Wanandi SI. Correlation between hypoxia inducible factor -1α and renin expression in rats kidney induced by cobalt chloride. Med J Indones. 2012; 21(3): 128-32.
26. Cheong HI, Asosingh K, Stephens OR, Queisser KA, Xu W, Willard B, et al. Hypoxia sensing through β-adrenergic receptors. JCI Insight. 2016; 1(21): e90240.
27. Coles JP. Regional ischemia after head injury. Current Opinion in Critical Care. 2004; 10(2):120-5.
28. Ding Z, Zhang J, Xu J, Sheng G, Huang G. Propofol administration modulates AQP-4 expression and brain edema after traumatic brain injury. Cell Biochem Biophys. 2013; 67(2): 615-22.
29. Thelin EP, Frostell A, Mulder J, Mitsios N, Damberg P, Aski SN, et al. Lesion size is exacerbated in hypoxic rats whereas hypoxia-inducible factor-1 alpha and vascular endothelial growth factor increase in injured normoxic rats: A prospective cohort study of secondary hypoxia in focal traumatic brain injury. Front Neurol. 2016; 7: 23.
30. Kasivisvanathan V, Shalhoub J, S. Lim C, C. Shepherd A, Thapar A, H. Davies A. Hypoxia-Inducible Factor-1 in Arterial Disease: A Putative Therapeutic Target. Curr Vasc Pharmacol. 2011; 15(5): 371-4.
31. Khan M, Im Y Bin, Shunmugavel A, Gilg AG, Dhindsa RK, Singh AK, et al. Administration of S-nitrosoglutathione after traumatic brain injury protects the neurovascular unit and reduces secondary injury in a rat model of controlled cortical impact. J Neuroinflammation. 2009; 6: 32.