Published in final edited form as:
Respir Physiol Neurobiol. 2008 December 10; 164(1-2): 282–287. doi:10.1016/j.resp.2008.07.017.

HIF-1 and ventilatory acclimatization to chronic hypoxia

Frank L. Powell and **Zhenxing Fu**
Department of Medicine and White Mountain Research Station University of California, San Diego La Jolla, CA 92093-0623 USA

Abstract

Ventilatory acclimatization to hypoxia (VAH) is a time-dependent increase in ventilation and ventilatory O_2-sensitivity that involves plasticity in carotid body chemoreceptors and CNS respiratory centers. Hypoxia inducible factor-1α (HIF-1α) controls the expression of several genes that increase physiological O_2 supply. Studies using transgenic mice show HIF-1α expression in the carotid bodies and CNS with chronic sustained and intermittent hypoxia is important for VAH. Other O_2-sensitive transcription factors such as HIF-2α may be important for VAH by reducing metabolic O_2 demands also. Specific gene targets of HIF-1α shown to be involved in VAH include erythropoietin, endothelin-1, neuronal nitric oxide synthase and tyrosine hydroxylase. Other HIF-1α targets that may be involved in VAH include vascular endothelial growth factor, heme oxygenase 1 and cytoglobin. Interactions between these multiple pathways and feedback control of HIF-1α expression from some of the targets support a complex and powerful role for HIF-1α in neural plasticity of physiological control circuits with chronic hypoxia.

Keywords

carotid body; endothelin-1; erythropoietin; gene expression; heme oxygenase; neural plasticity; nitric oxide; transgenic mice; vascular endothelial growth factor

1. Introduction

Ventilatory acclimatization to hypoxia (VAH) is a time-dependent increase in ventilation that occurs in humans and animals exposed to chronic hypoxia (Powell et al., 1998). VAH involves an increase in the acute hypoxic ventilatory response (HVR) that is most clearly demonstrated when hypocapnia is prevented during hypoxic hyperventilation, i.e. isocapnic hypoxia (Powell et al., 2000). The increased HVR results from both enhanced carotid body O_2-sensitivity (Bisgard, 2000) and plasticity in CNS integration and processing of arterial chemoreceptor afferent input (Dwinell and Powell, 1999). Another feature of plasticity in ventilatory control with chronic hypoxia is ventilatory deacclimatization to hypoxia (VDH), which is a persistent hyperventilation when normoxia is restored after chronic hypoxia (Powell et al., 1998). The mechanisms for VDH are unknown but it cannot be simply explained by a metabolic acidosis in the cerebrospinal fluid stimulating ventral medullary CO_2-sensitive chemoreceptors as originally hypothesized in the 1960s (Dempsey and Forster, 1982). VAH and VDH require

Corresponding author: Frank L. Powell, Ph.D., Division of Physiology, Department of Medicine, University of California San Diego, 9500 Gilman Drive, #0623A, La Jolla, CA 92093-0623, USA, Phone: 858-534-4191, Fax: 858-534-4812, Email: E-mail: fpowell@ucsd.edu.

hours to days of hypoxic exposure so it is reasonable to hypothesize they involve changes in gene expression in the neural circuits controlling breathing. A likely factor controlling changes in the expression of genes involved in VAH is Hypoxia Inducible Factor 1 (HIF-1), which was discovered as an important transcription factor for erythropoietin (EPO) and has been described as a master regulator for O_2 homeostasis (Semenza, 1998). Since this discovery, HIF-1 has been shown to increase expression of a wide variety of genes besides EPO that are involved in O_2 delivery and metabolism (Semenza, 1999).

1.1. HIF-1 molecular biology and physiology

This topic has been covered in detail in other recent reviews (e.g. Weidemann and Johnson, 2008). HIF-1 induces physiological responses to hypoxia by binding to an 8 base pair hypoxia response element (HRE) in hypoxia responsive genes and activating transcription. HIF-1 activity depends on phosphorylation and its binding in the DNA major groove is redox-sensitive. HIF-1 is a heterodimer composed of HIF-1α and HIF-1β. HIF-1β is also known as the aryl hydrocarbon receptor nuclear translocator, ARNT. Both HIF-1 subunits are expressed constitutively but HIF-1α is subject to degradation under normoxic conditions. Prolyl hydroxylases (PHDs) act on two specific prolyl residues in the oxygen-dependent-degradation domains (ODD) of HIF-1α subunits in the presence of oxygen, Fe^{2+} and 2-oxogluterate. Under normoxic conditions, PHD is hydroxylated and the ODD binds the Von Hippel-Lindau protein. This provides recognition for the E3 ubiquitin ligase complex and leads to proteasomal degradation of HIF-1α. Other hydroxylases modify asparagine residues in the carboxy-terminal-trans-activation domain of HIF-1α and modulate HIF-1α activity instead of stability in response to oxygen.

The first correlation demonstrated between a physiological stimulus, increased HIF-1 and an adaptive physiological response was increased vascular endothelial growth factor (VEGF) expression and increased cardiac vascularization in fetal lambs subjected to chronic anemia (Martin et al., 1998). HIF-1α is also essential for vascularization during normal embryonic development and homozygous deletion of the HIF-1α gene in mice is embryonic lethal (Ryan et al., 1998). Although the regression of blood vessels and massive cell death in cephalic mesenchyme of homozygous HIF-1α knockout mice is similar to that of homozygous VEGF knockout mice, homozygous HIF-1α knockout mice actually show increased VEGF (reviewed by Semenza, 1999). Therefore, HIF-1α exerts important developmental effects by other non-VEGF pathways too. HIF-1 gene products that are relevant to ventilatory acclimatization to hypoxia are considered in Section 5.

1.2. Other O_2-sensitive Transcription Factors

Besides HIF-1α, other transcription factors in the basic helix-loop-helix/PAS superfamily include HIF-2α and HIF-3α. HIF-2α is also referred to as endothelial PAS domain protein-1 (EPAS-1) and like HIF-1α, it is stabilized by hypoxia (Wiesener et al., 1998). Gene expression studies show relatively few target genes for HIF-2α versus HIF-1α and even fewer of these are uniquely controlled by HIF-2α (Warnecke et al., 2008). The effects of HIF-1α knockdown are generally larger than those of HIF-2α knockdown on genes controlled by both factors (Warnecke et al., 2008). However, a notable exception is EPO and there is evidence that HIF-2α may be the most important factor controlling erythropoiesis in hypoxia, at least in mice (Gruber et al., 2007).

Involvement of HIF-2α in VAH remains to be demonstrated but it appears likely. EPO is produced in the brain and is important for CNS mechanisms of VAH as described later. The effect of HIF-2α on EPO in the brain is not known yet but is reasonable to hypothesize that it may be stimulatory, as it is for systemic EPO (Gurber et al., 2007). A role for HIF-2α in carotid body mechanisms of VAH has also been suggested, as discussed later. Finally HIF-2α appears

to be more important than HIF-1α in metabolic reprogramming in skeletal muscle. Skeletal muscle from mice with the PHd1 gene deleted, which prevents normal degradation of HIF-1α in normoxia, shows decreased O_2 consumption and shift towards anaerobic glycolysis (Aragones et al., 2008). HIF expression patterns indicated that HIF-2α is more important than HIF-1α in these metabolic changes to reduce O_2 demand under hypoxic conditions. Such decreased O_2 consumption complements the physiological effects of ventilatory acclimatization to increase O_2 supply in chronic hypoxia.

The physiological role of HIF-3α is not clear yet (Heidelbreder et al., 2003). However, descriptive studies show similar patterns for expression of HIF-2α and HIF-3α in chronic, sustained versus intermittent hypoxia (Lam et al., 2008b), which suggests the functions similar to those discussed above for HIF-2α.

Hypoxia activates several other transcription factors besides HIF but in general their role in VAH is not known (Cummins and Taylor, 2005). Nuclear factor-kappaB (NF-κB) is a master transcriptional regulator of inflammatory and antiapoptotic gene expression that is also increased by hypoxia (Cummins et al., 2007). Chronic hypoxia increases inflammatory cytokines in the carotid body that are known to activate NF-κB and increase calcium influx to glomus cells during acute hypoxia (Lam et al., 2008a). Activator protein-1 (AP-1) is another hypoxia sensitive transcription factor that appears to be involved in carotid body mechanisms of acclimatization to chronic intermittent hypoxia (Prabhakar and Kumar, 2004).

2. HIF-1α and the Hypoxic Ventilatory Response (HVR)

HIF-1α is not necessary for a normal HVR in mice but it does contribute to O_2-sensing in the carotid body. Kline and co-workers (2002) studied transgenic mice with a heterozygous loss of function in the HIF-1α gene and found no difference in carotid body morphology, the ventilatory response to CO_2, or the acute HVR compared to control, wild type mice. However, carotid bodies in these heterozygous HIF-1α knock out mice had a depressed neural response to hypoxia. The investigators hypothesize that non-carotid body arterial chemoreceptors compensate for a loss of carotid body function to preserve a normal HVR, similar to what has been observed in carotid body denervation experiments. Their theory was supported by results showing a depressed HVR in heterozygous HIF-1α knockout mice following vagotomy, which removed sensory input from non-carotid body arterial chemoreceptors. The authors speculate that non-carotid body chemoreceptors do not depend on HIF-1α for normal O_2-sensitivity but plasticity in CNS integration of arterial chemoreceptor afferent input could also explain the results.

The mechanism for depressed O_2-sensitivity in the carotid bodies of heterozygous HIF-1α knockout mice is unknown. The carotid body response to cyanide is not depressed in the heterozygous HIF-1α knockout mice, which indicates that histotoxic versus hypoxic chemosensory mechanisms are under differential genetic control. Lahiri and co-workers (Roy et al., 2007) have suggested that HIF-1α and neural activity in the carotid body depend on a common O_2-sensor that is mitochondrial cytochrome a_3. However, it is not obvious how this could be involved in the depressed O_2-sensitivity in the carotid bodies of heterozygous HIF-1α knock out mice.

3. HIF-1α and Ventilatory Acclimatization to Hypoxia (VAH)

3.1. Arterial chemoreceptor plasticity

HIF1-α increases in the carotid body after as little as 1 h of hypoxia (Kline et al., 2002; Pascual et al., 2001). The heterozygous HIF1-α knockout mice discussed above also show abnormal VAH (Kline et al., 2002). Chronic hypoxia increased the HVR in wild type mice, primarily by

an effect on respiratory frequency (fR), but fR actually decreased in hypoxia after acclimatization in the heterozygous HIF1-α knockout mice. Acute hypoxia increased carotid body activity less in heterozygous HIF1-α knockout mice compared to normal mice so this depression of carotid body O_2-sensitivity likely contributed to the depressed VAH. However, carotid body O_2-sensitivity has not been recorded after chronic hypoxia in the heterozygous HIF1-α knockout mice yet and CNS mechanisms of VAH may be compromised in these transgenic mice also (see below).

3.2. Central Nervous System (CNS) plasticity

In addition to increased carotid body O_2-sensitivity with chronic hypoxia, VAH involves an increase in the CNS gain of the HVR, which is defined as an increased ventilatory motor response to a given afferent input from carotid bodies. This has been demonstrated in anesthetized rats as an increased phrenic nerve response to electrical stimulation of the carotid sinus nerve after chronic hypoxia (Dwinell and Powell, 1999). HIF-1α increases in the CNS respiratory centers after as little as 1 hour and peak changes in neurons are observed after 6 hours (Pascual et al., 2001). To investigate the role of HIF-1α in the increased CNS gain of the HVR with chronic hypoxia, we have begun to study transgenic mice with a CNS-specific deletion of the HIF-1α gene (Helton et al., 2005).

Preliminary results show the acute HVR is normal in CNS HIF-1α knockout mice. However in contrast to wild type mice, the knockout mice do not show an increased HVR after chronic hypoxia (Bavis et al., 2007). The HIF-1α gene was not deleted in the carotid bodies of these CNS-specific knockout mice (Kim, Fu and Powell, unpublished observations) indicating that HIF-1α in the CNS also plays a role in VAH. Determining the exact sites of plasticity that are induced by HIF-1α in the CNS and contribute to VAH will require spatial and temporal resolution not possible with the transgenic breeding strategies used for these studies.

4. HIF-1α and Chronic Intermittent Hypoxia (CIH)

CIH has been studied as a model for sleep disordered breathing and obstructive sleep apnea and consists of repeated bouts of hypoxia lasting seconds to minutes for several hours per day. Peng and colleagues (2006) studied the role of HIF-1α in ventilatory control plasticity during CIH using transgenic mice with one HIF-1α gene deleted (heterozygous HIF-1α knockouts). In control, wild type mice, CIH (nine 15 second bouts of hypoxia per hour, 8 hrs per day, 10 days) caused changes in ventilation and the poikilocapnic HVR similar to what is observed with chronic sustained hypoxia, i.e. ventilation increased in hypoxia and normoxia. However, CIH also causes changes that are not observed with chronic sustained hypoxia. For example, CIH pretreatment enabled long term facilitation of the carotid body response (sensory LTF; Peng and Prabhakar, 2004), and enhanced long term facilitation of phrenic nerve activity following acute intermittent hypoxia (phrenic LTF; LING et al., 2001). In heterozygous HIF-1α knockout mice, all of these responses to CIH were either absent or attenuated indicating a necessary role for HIF-1α in intermittent as well as continuous chronic hypoxia.

Results of reactive oxygen species (ROS) measurements and experiments with ROS scavengers suggested the effects of CIH involve positive feedback interactions between HIF-1α and ROS. Experiments using PC12 cells as a model for carotid body glomus cells suggest that intermittent hypoxia induces HIF-1α transcriptional activity via a novel signaling pathway involving calcium/calmodulin-dependent (CaM) kinase II (Yuan et al., 2005). Determining the relationships between control of HIF-1α expression and its effects in intermittent versus sustained hypoxia should prove useful in unraveling the mechanisms of ventilatory acclimatization that are unique to these two patterns of chronic hypoxia.

5. Potential mechanisms for plasticity induced by HIF-1α

This section considers the targets of HIF-1α that could contribute to VAH. Metabolic responses to HIF could also contribute to VAH by altering local O_2 levels that are the stimuli for hypoxic responses, or by modulating metabolic control of neurotransmission. However, such potential effects are not considered further in this review.

5.1. Erythropoietin (EPO)

HIF-1α was originally discovered as an important transcription factor for EPO, which is released from the kidneys in response to hypoxia and increases the oxygen carrying capacity of blood by stimulating erythropoiesis in bone marrow (Semenza, 1999). However, EPO and its receptor (EPO-R) have also been localized to the CNS in glia and neurons where it must have different functions. EPO in the brain is reported to have a protective role during hypoxemic or ischemic injury (Gassmann et al., 2003). Recent studies with transgenic mice that overexpress human EPO in the brain support a role for EPO in VAH also (Soliz et al., 2005). The Tg21 line of transgenic mice have over 3-fold higher EPO levels in the brainstem than wild type mice but plasma EPO and hematocrits are not elevated. With chronic hypoxia, Tg21 mice increase ventilation more than control, wild type mice, although the persistent hyperventilation when normoxia is restored is similar in both strains. EPO-R is localized in respiratory centers such as pre-Bötzinger complex, the nucleus tractus solitarius (NTS) and catecholaminergic cell groups in the medulla (A1C1, A2C2) and pons (A5, A6). The soluble EPO receptor (sEPO-R) may play the primary role in EPO effects on VAH in the brain (Soliz et al., 2007). sEPO-R in the brain is downregulated by chronic hypoxia and intracerebroventricular infusion of sEPO-R to chronically hypoxic mice decreases EPO and reverses VAH. Hence, HIF-1α affects both hematological and ventilatory acclimatization to chronic hypoxia.

Studies on a different transgenic mouse (Tg6), which over-expresses EPO in the brain and systemically so hematocrit increases, suggest that EPO exerts effects on the HVR both centrally and peripherally (Soliz et al., 2007). In Tg6 mice, norepinepherine is increased in the A5 cell group of the pons and this is known to increase respiratory frequency (Dick and Coles, 2000). Systemic EPO also increases frequency by actions on the carotid body (Soliz et al., 2005).

5.2. Vascular Endothelial Growth Factor (VEGF)

VEGF is one of the most studied HIF-1α targets because of its role in increasing tumor vascularity, which suggests a promising avenue for anticancer therapeutics. However, VEGF has been localized also to motor neurons (Murakami et al., 2003) and may play a role in neural plasticity during chronic hypoxia. VEGF activates similar cellular signaling pathways as brain derived neurotrophic growth factor (BDNF), which has been shown to play a critical role in long term facilitation of ventilation (LTF) following intermittent hypoxia by binding TrkB receptors (Baker-Herman et al., 2004). VEGF effects are exerted by binding another tyrosine kinase receptor, VEGFR2 (Roitbak et al., 2008). Also, VEGF and BDNF are both involved in neural plasticity after cerebral ischemia (Chen et al., 2005; Roitbak et al., 2008). A similar role for VEGF inducing neural plasticity in respiratory control circuits during chronic hypoxia could support the other effects of HIF-1α on acclimatization but it remains to be demonstrated experimentally.

5.3. Endothelin-1 (ET-1)

ET-1 is a vasoactive peptide that increases with chronic hypoxia in the carotid body (J. Chen et al., 2002; Lam et al., 2008b) as well as other O_2-sensitive tissues such as the pulmonary artery (Li et al., 1994). ET-1 is also widely distributed in the brain where it plays an important

role in the response to stress (Kurihara et al., 2000). Hypoxia regulates ET-1 expression through HIF-1α (Hu et al., 1998) and a specific role for ET-1 in mediating VAH is supported by experiments showing that antagonists for ET_A receptors reverse the increased O_2-sensitivity observed in carotid bodies from chronically hypoxic rats (Y. Chen et al., 2002). While ET-1 alone does not excite carotid bodies, it does enhance the response to hypoxia and this effect is mediated by ET_A receptors on glomus cells (J. Chen et al., 2002), which are increased with chronic hypoxia (Y. Chen et al., 2002). ET-1 may increase O_2-sensitivity of glomus cells by effects on the G-coupled ET_A receptor to increase calcium currents (Y. Chen et al., 2002). However, ET-1 may also have effects on CNS mechanisms of hypoxia responses considering that the HVR is blunted in anesthetized mice with a heterozygous knock-out of the ET-1 gene (Kuwaki et al., 1996). Experiments manipulating ET-1 and ET_A expression specifically in the CNS and during acclimatization to chronic hypoxia (versus life-long deletion) will be necessary to test this hypothesis.

5.4. Heme oxygenase (HO)

Two isoforms of HO may be involved in ventilatory O_2-sensitivity via O_2-dependent catalysis of heme into CO, biliverdin and iron (Maines and Gibbs, 2005) and both forms affect neuronal ion channels (Boehning and Snyder, 2003; Lopez-Barneo et al., 2004). HO-1 is known to be induced by HIF-1α (Semenza, 1999) and HO-1 is also induced by chronic hypoxia in the rostral ventrolateral medulla, where it has been hypothesized to increase central O_2-sensitivity (Mazza et al., 2001). However, it should be noted that HO-1 may be activated by more stimuli than any other gene (Maines and Gibbs, 2005) so the changes observed in chronic hypoxia may not depend on HIF-1α. Also, a specific role for HO-1 in central O_2-sensitivity remains to be demonstrated.

HO-2 is a constitutive isoform that is also expressed in the rostral ventrolateral medulla, the C1 region and pre-Bötzinger complex, all of which demonstrate O_2-sensitivity and affect breathing (Mazza et al., 2001). HO-2 is hypothesized to be involved in acute ventilatory responses to CNS hypoxia but levels do not change with chronic hypoxia as expected if it was important for plasticity (Mazza et al., 2001; Adachi et al., 2004).

5.5. Tyrosine hydroxylase (TH)

Dopaminergic modulation of the HVR in both the CNS and arterial chemoreceptor changes with ventilatory acclimatization to hypoxia (Huey et al., 2003). TH is the rate limiting enzyme for biosynthesis of dopamine and its expression is increased by hypoxia. However, the role of HIF-1α in causing changes in TH and dopamine during chronic hypoxia is not certain. The HRE in the TH gene can bind HIF-1α but AP1 appears to be the main factor responsible for increased TH gene expression with hypoxia in PC 12 cells (Millhorn et al., 1997). Both TH transcription and mRNA stability are increased by hypoxia in PC12 cells (Czyzyk-Krzeska et al., 1994). Hence, HIF-1α may be involved in VAH through effects on TH but this has not been proven conclusively.

In the medulla, HIF-1α is increased by hypoxia selectively in cardiorespiratory centers and is co-localized with TH in many-- but not all -- catecholaminergic neurons, e.g. A1C1 and A2C2 cell groups (Pascual et al., 2001). In carotid body cells, HIF-2α correlates more closely than HIF-1α with increased TH in chronic hypoxia. Both chronic intermittent and sustained hypoxia increase TH and HIF-2α but HIF-1α is only increased by chronic sustained hypoxia (Lam et al., 2008b). HIF-1α in the carotid body may be more important for hypertrophy and increased vascularization than changes in O_2-sensitivity with chronic sustained hypoxia. On the other hand, HIF-1α correlates more closely than HIF-2α with developmental changes in carotid body TH and O_2-sensitivity. HIF-1α decreases in the carotid body as O_2-sensitivity increases during the perinatal period, while HIF-2α does not change during the same period (Roux et al.,

2005). Experimental interventions instead of correlative studies will be necessary to determine the role of various HIFs in controlling TH and mechanisms of ventilatory acclimatization to hypoxia.

5.6. Nitric Oxide Synthase (NOS)

Generally, HIF-1α is thought to affect nitric oxide (NO) through induction of inducible NOS (iNOS, Semenza, 1999) but recent experiments show that hypoxia leads to activation of a novel neuronal NOS (nNOS) promoter. Ward and co-workers (2005) showed that HIF-1α binds specifically under hypoxic conditions to a HRE found in the human nNOS sequence and produces mRNA with high translational efficiency. Nitric oxide (NO) itself has important effects on ventilation and the HVR. Increasing NO in the nucleus tractus solitarius stimulates ventilation (Ogawa et al., 1995) while increasing NO in the carotid body depresses O_2-sensitivity and thereby the HVR (Bisgard, 2000). Hence, changes in NO from induction of nNOS during chronic hypoxia could be involved in ventilatory acclimatization.

Hypoxemia from anemia increases HIF-1α and mRNA for nNOS in the cerebral cortex (McLaren et al., 2007) and chronic hypoxia increases mRNA and protein for nNOS in both peripheral and central neurons (Prabhakar et al., 1996). Selective inhibition nNOS in the brainstem has no effect on ventilation or the HVR in control mice but it decreases ventilation in chronically hypoxic mice (Hasnaoui-Saadani et al., 2007). Hence, effects of HIF-1α on NO appear to enhance O_2 delivery from effects on ventilation in chronic hypoxia, as well as from the effects of NO-induced vasodilation on vascular O_2 delivery. NO also decreases O_2 demand by its inhibitory effects mitochondrial respiration. Ward and colleagues (2005) point out that this latter effect could establish negative feedback control of nNOS expression at the transcriptional level by NO-dependent prolyl hydroxylase disinhibition that prevents HIF-1α stabilization.

A further complexity in nNOS regulation by HIF-1α may involve ET-1, another HIF-1α target involved in VAH (see above). In the kidney, ET-1 upregulates nNOS through an effect on ET_A receptors (Cummins et al., 2007). If ET-1 has the same effect on nNOS in the brain or carotid bodies, then it could provide another level of interaction between HIF-1α and nNOS in VAH.

5.7. Neuroglobin (Ngb) and Cytoglobin (Cygb)

Genomic studies lead to the discovery of Ngb and Cygb, which bind oxygen reversibly via iron atoms in a porphyrin ring in a fashion similar to hemoglobin and myoglobin. Ngb is a candidate for mediating neural plasticity in hypoxic neurons because it is up-regulated in the brain by hypoxic-ischemic injury, it protects neurons from further damage by hypoxia, it may scavenge reactive oxygen species and NO, and it may function as an oxygen sensor (Sun et al., 2001; Fordel et al., 2004). However, Ngb expression does not depend on HIF-1α (Fordel et al., 2004).

In contrast, increased Cygb expression in hypoxia does depend on HIF-1α and comparative studies plus the O_2 binding properties of Cygb suggest it may facilitate O_2 delivery in the hypoxic tissue. Hence, HIF-1α could enhance O_2 delivery in the CNS serially: firstly, by its effect to increase hemoglobin and arterial O_2 content (via EPO), and secondly, by increased Cygb to facilitate tissue O_2 delivery (similar to the role of myoglobin in skeletal muscle) (Williams et al., 2008). This could modulate ventilatory acclimatization to hypoxia by causing time-dependent changes in the P_{O_2} at O_2-chemosensitive sites in the brain.

6. Conclusions and Future Directions

HIF-1α produces a suite of coordinated responses to chronic hypoxia. These contribute to ventilatory acclimatization to hypoxia but they cannot explain every aspect of the plasticity observed in ventilatory control with chronic hypoxia. Also, the effects of HIF-1α differ between chronic intermittent versus sustained hypoxia. Together, these observations suggest that experimental manipulations of HIF-1α in specific pathways of ventilatory acclimatization may help us dissect the integrated response to various patterns of hypoxia. Understanding how the individual mechanisms interact could help us develop therapeutics for treating specific patterns of hypoxemia, for example intermittent hypoxia with obstructive sleep apnea versus sustained hypoxia with chronic obstructive pulmonary disease.

Acknowledgments

This work was supported by NIH 1R01HL081823 and the University of California White Mountain Research Station.

References

Adachi T, Ishikawa K, Hida W, Matsumoto H, Masuda T, Date F, Ogawa K, Takeda K, Furuyama K, Zhang YZ, Kitamuro T, Ogawa H, Maruyama Y, Shibahara S. Hypoxemia and blunted hypoxic ventilatory responses in mice lacking heme oxygenase-2. Biochemical and Biophysical Research Communications 2004;320:514–522. [PubMed: 15219859]

Aragones J, Schneider M, van Geyte K, Fraisl P, Dresselaers T, Mazzone M, Dirkx R, Zacchigna S, Lemieux H, Jeoung NH, Lambrechts D, Bishop T, Lafuste P, Diez-Juan A, Harten SK, Van Noten P, De Bock K, Willam C, Tjwa M, Grosfeld A, Navet R, Moons L, Vandendriessche T, Deroose C, Wijeyekoon B, Nuyts J, Jordan B, Silasi-Mansat R, Lupu F, Dewerchin M, Pugh C, Salmon P, Mortelmans L, Gallez B, Gorus F, Buyse J, Sluse F, Harris RA, Gnaiger E, Hespel P, Van Hecke P, Schuit F, Van Veldhoven P, Ratcliffe P, Baes M, Maxwell P, Carmeliet P. Deficiency or inhibition of oxygen sensor Phd1 induces hypoxia tolerance by reprogramming basal metabolism. Nat Genet 2008;7:170–180. [PubMed: 18176562]

Baker-Herman TL, Fuller DD, Bavis RW, Zabka AG, Golder FJ, Doperalski NJ, Johnson RA, Watters JJ, Mitchell GS. BDNF is necessary and sufficient for spinal respiratory plasticity following intermittent hypoxia. Nat Neurosci 2004;7:48–55. [PubMed: 14699417]

Bavis RW, Powell FL, Bradford A, Hsia CCW, Peltonen JE, Soliz J, Zeis B, Fergusson EK, Fu Z, Gassmann M, Kim CB, Maurer J, McGuire M, Miller BM, O'Halloran KD, Paul RJ, Reid SG, Rusko HK, Tikkanen HO, Wilkinson KA. Respiratory plasticity in response to changes in oxygen supply and demand. Integrative and Comparative Biology 2007;47:532–551.

Bisgard GE. Carotid body mechanisms in acclimatization to hypoxia. Respir Physiol 2000;121:237–246. [PubMed: 10963778]

Boehning D, Snyder SH. Novel neural modulators. Annu Rev Neurosci 2003;7:105–131. [PubMed: 14527267]

Chen J, He L, Dinger B, Stensaas L, Fidone S. Role of endothelin and endothelin Atype receptor in adaptation of the carotid body to chronic hypoxia. Am J Physiol Lung Cell Mol Physiol 2002;282:L1314–L1323. [PubMed: 12003788]

Chen J, Zhang C, Jiang H, Li Y, Zhang L, Robin A, Katakowski M, Lu M, Chopp M. Atorvastatin induction of VEGF and BDNF promotes brain plasticity after stroke in mice. J Cereb Blood Flow Metab 2005;25:281–290. [PubMed: 15678129]

Chen Y, Tipoe GL, Liong E, Leung S, Lam SY, Iwase R, Tjong YW, Fung ML. Chronic hypoxia enhances endothelin-1-induced intracellular calcium elevation in rat carotid body chemoreceptors and up-regulates ET_A receptor expression. Pflugers Arch 2002;443:565–573. [PubMed: 11907823]

Cummins EP, Taylor CT. Hypoxia-responsive transcription factors. Pflugers Arch 2005;450:363–371. [PubMed: 16007431]

Cummins EP, Comerford KM, Scholz C, Bruning U, Taylor CT. Hypoxic regulation of NF-κB signaling. Methods Enzymol 2007;7:479–492. [PubMed: 17998070]

Czyzyk-Krzeska MF, Furnari BA, Lawson EE, Millhorn DE. Hypoxia increases rate of transcription and stability of tyrosine hydroxylase mRNA in pheochromocytoma (PC12) cells. J Biol Chem 1994;7:760–764. [PubMed: 7903970]

Dempsey JA, Forster HV. Mediation of ventilatory adaptations. Physiol Rev 1982;7:262–346. [PubMed: 6798585]

Dick TE, Coles SK. Ventrolateral pons mediates short-term depression of respiratory frequency after brief hypoxia. Respir Physiol 2000;121:87–100. [PubMed: 10963767]

Dwinell MR, Powell FL. Chronic hypoxia enhances the phrenic nerve response to arterial chemoreceptor stimulation in anesthetized rats. J Appl Physiol 1999;7:817–823. [PubMed: 10444644]

Fordel E, Geuens E, Dewilde S, De Coen W, Moens L. Hypoxia/Ischemia and the Regulation of Neuroglobin and Cytoglobin Expression. IUBMB Life 2004;56:681–687. [PubMed: 15804832]

Gassmann M, Heinicke K, Soliz J, Ogunshola OO. Non-erythroid functions of erythropoietin. Adv Exp Med Biol 2003;7:323–330. [PubMed: 14713131]

Gruber M, Hu CJ, Johnson RS, Brown EJ, Keith B, Simon MC. Acute postnatal ablation of Hif-2α results in anemia. Proc Natl Acad Sci U S A 2007;104:2301–2306. [PubMed: 17284606]

Hasnaoui-Saadani R, Alayza RC, Launay T, Pichon A, Quidu P, Beaudry M, Leon-Velarde F, Richalet JP, Duvallet A, Favret F. Brain stem NO modulates ventilatory acclimatization to hypoxia in mice. J Appl Physiol 2007;103:1506–1512. [PubMed: 17690195]

Heidbreder M, Frohlich F, Johren O, Dendorfer A, Qadri F, Dominiak P. Hypoxia rapidly activates HIF-3α mRNA expression. FASEB J 2003;17:1541–1543. [PubMed: 12824304]

Helton R, Cui J, Scheel JR, Ellison JA, Ames C, Gibson C, Blouw B, Ouyang L, Dragatsis I, Zeitlin S, Johnson RS, Lipton SA, Barlow C. Brain-specific knock-out of hypoxia-inducible factor-1α reduces rather than increases hypoxic-ischemic damage. J Neurosci 2005;7:4099–4107. [PubMed: 15843612]

Hu J, Discher DJ, Bishopric NH, Webster KA. Hypoxia regulates expression of the endothelin-1 gene through a proximal hypoxia-inducible factor-1 binding site on the antisense strand. Biochem Biophys Res Commun 1998;7:894–899. [PubMed: 9588211]

Huey KA, Szewczak JM, Powell FL. Dopaminergic mechanisms of neural plasticity in respiratory control: transgenic approaches. Respir Physiol Neurobiol 2003;7:133–144. [PubMed: 12809614]

Kline DD, Peng YJ, Manalo DJ, Semenza GL, Prabhakar NR. Defective carotid body function and impaired ventilatory responses to chronic hypoxia in mice partially deficient for hypoxia-inducible factor 1 α. Proc Natl Acad Sci U S A 2002;99:821–826. [PubMed: 11792862]

Kurihara Y, Kurihara H, Morita H, Cao WH, Ling GY, Kumada M, Kimura S, Nagai R, Yazaki Y, Kuwaki T. Role of endothelin-1 in stress response in the central nervous system. Am J Physiol Regul Integr Comp Physiol 2000;279:R515–R521. [PubMed: 10938240]

Kuwaki T, Cao WH, Kurihara Y, Kurihara H, Ling GY, Onodera M, Ju KH, Yazaki Y, Kumada M. Impaired ventilatory responses to hypoxia and hypercapnia in mutant mice deficient in endothelin-1. Am J Physiol 1996;270:R1279–R1286. [PubMed: 8764295]

Lam SY, Tipoe GL, Liong EC, Fung ML. Chronic hypoxia upregulates the expression and function of proinflammatory cytokines in the rat carotid body. Histochem Cell Biol. 2008a

Lam SY, Tipoe GL, Liong EC, Fung ML. Differential expressions and roles of hypoxia-inducible factor-1α, -2α and -3α in the rat carotid body during chronic and intermittent hypoxia. Histol Histopathol 2008b;23:271–280. [PubMed: 18072084]

Li H, Chen SJ, Chen YF, Meng QC, Durand J, Oparil S, Elton TS. Enhanced endothelin-1 and endothelin receptor gene expression in chronic hypoxia. J Appl Physiol 1994;77:1451–1459. [PubMed: 7836152]

Lopez-Barneo J, del Toro R, Levitsky KL, Chiara MD, Ortega-Saenz P. Regulation of oxygen sensing by ion channels. J Appl Physiol 2004;96:1187–1195. [PubMed: 14766769]

Maines MD, Gibbs PE. 30 some years of heme oxygenase: from a “molecular wrecking ball” to a “mesmerizing” trigger of cellular events. Biochem Biophys Res Commun 2005;7:568–577. [PubMed: 16183036]

Martin C, Yu AY, Jiang BH, Davis L, Kimberly D, Hohimer AR, Semenza GL. Cardiac hypertrophy in chronically anemic fetal sheep: Increased vascularization is associated with increased myocardial

expression of vascular endothelial growth factor and hypoxia-inducible factor 1. Am J Obstet Gynecol 1998;7:527–534. [PubMed: 9539521]

Mazza E, Thakkar-Varia S, Tozzi CA, Neubauer JA. Expression of heme oxygenase in the oxygen-sensing regions of the rostral ventrolateral medulla. J Appl Physiol 2001;7:379–385. [PubMed: 11408455]

McLaren AT, Marsden PA, Mazer CD, Baker AJ, Stewart DJ, Tsui AK, Li X, Yucel Y, Robb M, Boyd SR, Liu E, Yu J, Hare GM. Increased expression of HIF-1α, nNOS, and VEGF in the cerebral cortex of anemic rats. Am J Physiol Regul Integr Comp Physiol 2007;292:R403–R414. [PubMed: 16973934]

Millhorn DE, Raymond R, Conforti L, Zhu W, Beitner-Johnson D, Filisko T, Genter MB, Kobayashi S, Peng M. Regulation of gene expression for tyrosine hydroxylase in oxygen sensitive cells by hypoxia. Kidney Int 1997;7:527–535. [PubMed: 9027733]

Murakami T, Ilieva H, Shiote M, Nagata T, Nagano I, Shoji M, Abe K. Hypoxic induction of vascular endothelial growth factor is selectively impaired in mice carrying the mutant SOD1 gene. Brain Res 2003;7:231–237. [PubMed: 14556945]

Ogawa H, Mizusawa A, Kikuchi Y, Hida W, Miki H, Shirato K. Nitric oxide as a retrograde messenger in the nucleus tractus solitarii of rats during hypoxia. J Physiol 1995;7:495–504. [PubMed: 7473213]

Pascual O, Denavit-Saubie M, Dumas S, Kietzmann T, Ghilini G, Mallet J, Pequignot JM. Selective cardiorespiratory and catecholaminergic areas express the hypoxia-inducible factor-1α (HIF-1α) under *in vivo* hypoxia in rat brainstem. Eur J Neurosci 2001;14:1981–1991. [PubMed: 11860493]

Peng YJ, Prabhakar NR. Effect of two paradigms of chronic intermittent hypoxia on carotid body sensory activity. J Appl Physiol 2004;96:1236–1242. [PubMed: 14660510]

Peng YJ, Yuan G, Ramakrishnan D, Sharma SD, Bosch-Marce M, Kumar GK, Semenza GL, Prabhakar NR. Heterozygous HIF-1α deficiency impairs carotid body-mediated systemic responses and reactive oxygen species generation in mice exposed to intermittent hypoxia. J Physiol 2006;577:705–716. [PubMed: 16973705]

Powell FL, Dwinell MR, Aaron EA. Measuring ventilatory acclimatization to hypoxia: comparative aspects. Respir Physiol 2000;7:271–284. [PubMed: 10967350]

Powell FL, Milsom WK, Mitchell GS. Time domains of the hypoxic ventilatory response. Respir Physiol 1998;7:123–134. [PubMed: 9716296]

Prabhakar NR, Kumar GK. Oxidative stress in the systemic and cellular responses to intermittent hypoxia. Biol Chem 2004;7:217–221. [PubMed: 15134334]

Prabhakar NR, Pieramici SF, Premkumar DR, Kumar GK, Kalaria RN. Activation of nitric oxide synthase gene expression by hypoxia in central and peripheral neurons. Brain Research Molecular Brain Research 1996;43:341–346. [PubMed: 9037552]

Roitbak T, Li L, Cunningham LA. Neural stem/progenitor cells promote endothelial cell morphogenesis and protect endothelial cells against ischemia via HIF-1α-regulated VEGF signaling. J Cereb Blood Flow Metab. 2008

Roux JC, Brismar H, Aperia A, Lagercrantz H. Developmental changes in HIF transcription factor in carotid body: relevance for O_2 sensing by chemoreceptors. Pediatr Res 2005;58:53–57. [PubMed: 15879294]

Roy A, Baby SM, Wilson DF, Lahiri S. Rat carotid body chemosensory discharge and glomus cell HIF-1 α expression in vitro: regulation by a common oxygen sensor. Am J Physiol Regul Integr Comp Physiol 2007;293:R829–R836. [PubMed: 17475674]

Ryan HE, Lo J, Johnson RS. HIF-1 α is required for solid tumor formation and embryonic vascularization. EMBO J 1998;7:3005–3015. [PubMed: 9606183]

Semenza GL. Hypoxia-inducible factor 1: master regulator of O_2 homeostasis. Curr Opin Genet Dev 1998;8:588–594. [PubMed: 9794818]

Semenza GL. Regulation of mammalian O_2 homeostasis by hypoxia-inducible factor 1. Annu Rev Cell Dev Biol 1999;15:551–578. [PubMed: 10611972]

Soliz J, Gassmann M, Joseph V. Soluble erythropoietin receptor is present in the mouse brain and is required for the ventilatory acclimatization to hypoxia. J Physiol 2007a;583:329–336. [PubMed: 17584830]

Soliz J, Soulage C, Hermann DM, Gassmann M. Acute and chronic exposure to hypoxia alters ventilatory pattern but not minute ventilation of mice overexpressing erythropoietin. Am J Physiol Regul Integr Comp Physiol 2007b;293:R1702–R1710. [PubMed: 17652365]

Soliz J, Joseph V, Soulage C, Becskei C, Vogel J, Pequignot JM, Ogunshola O, Gassmann M. Erythropoietin regulates hypoxic ventilation in mice by interacting with brainstem and carotid bodies. J Physiol 2005;568:559–571. [PubMed: 16051624]

Sun Y, Jin K, Mao XO, Zhu Y, Greenberg DA. Neuroglobin is up-regulated by and protects neurons from hypoxic-ischemic injury. Proc Natl Acad Sci U S A 2001;98:15306–15311. [PubMed: 11742077]

Ward ME, Toporsian M, Scott JA, Teoh H, Govindaraju V, Quan A, Wener AD, Wang G, Bevan SC, Newton DC, Marsden PA. Hypoxia induces a functionally significant and translationally efficient neuronal NO synthase mRNA variant. J Clin Invest 2005;115:3128–3139. [PubMed: 16276418]

Warnecke C, Weidemann A, Volke M, Schietke R, Wu X, Knaup KX, Hackenbeck T, Bernhardt W, Willam C, Eckardt KU, Wiesener MS. The specific contribution of hypoxia-inducible factor-2α to hypoxic gene expression in vitro is limited and modulated by cell type-specific and exogenous factors. Exp Cell Res. 2008

Weidemann A, Johnson RS. Biology of HIF-1α. Cell Death Differ 2008;7:621–627. [PubMed: 18259201]

Wiesener MS, Turley H, Allen WE, Willam C, Eckardt KU, Talks KL, Wood SM, Gatter KC, Harris AL, Pugh CW, Ratcliffe PJ, Maxwell PH. Induction of endothelial PAS domain protein-1 by hypoxia: characterization and comparison with hypoxia-inducible factor-1α. Blood 1998;92:2260–2268. [PubMed: 9746763]

Williams TM, Zavanelli M, Miller MA, Goldbeck RA, Morledge M, Casper D, Pabst DA, McLellan W, Cantin LP, Kliger DS. Running, swimming and diving modifies neuroprotecting globins in the mammalian brain. Proc Biol Sci 2008;7:751–758. [PubMed: 18089537]

Yuan G, Nanduri J, Bhasker CR, Semenza GL, Prabhakar NR. Ca^{2+}/calmodulin kinase-dependent activation of hypoxia inducible factor 1 transcriptional activity in cells subjected to intermittent hypoxia. J Biol Chem 2005;280:4321–4328. [PubMed: 15569687]