

Frontiers review
Time domains of the hypoxic ventilatory response

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Abstract

The ventilatory response to hypoxia depends on the pattern and intensity of hypoxic exposure and involves several physiological mechanisms. These mechanisms differ in their effect (facilitation or depression) on different components of ventilation (tidal volume and frequency) and in their time course (seconds to years). Some mechanisms last long enough to affect future ventilatory responses to hypoxia, indicating ‘memory’ or functional plasticity in the ventilatory control system. A standard terminology is proposed to describe the different time domains of the hypoxic ventilatory response (HVR) and to promote integration of results from different experimental preparations and laboratories. In general, the neurophysiological and neurochemical basis for short time domains of the HVR (seconds and minutes) are understood better than longer time domains (days to years), primarily because short time domains are studied in the laboratory more easily. Understanding the mechanisms for different time domains of the HVR has important implications for both basic and clinical science. © 1998 Elsevier Science B.V. All rights reserved.

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1. Introduction

The hypoxic ventilatory response (HVR) of vertebrates is not the product of a single mechanism,

but it is a complex interplay between several distinct mechanisms. Among the features distinguishing these different mechanisms are: (1) the specific stimuli that elicit them (e.g. pattern and intensity of hypoxic exposure); (2) the time course of the response (seconds to years); (3) the effects on the components of ventilation (tidal volume vs.

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frequency); (4) the direction of their effects (facilitation vs. depression); and (5) the neurochemicals necessary for their manifestation. Some mechanisms are sufficiently long-lasting to affect future ventilatory responses to hypoxia, indicating a degree of ‘memory’, or functional plasticity in the ventilatory control system. Apparently subtle differences in experimental protocols result in different combinations of these time-dependent mechanisms. Thus, in each specific circumstance, it is important to note which hypoxic ventilatory response is being elicited.

The primary goals of this short review are to summarize the different components of the HVR identified to date, and to compare and contrast their mechanisms. We also propose a common terminology, which hopefully will facilitate communication between investigators studying different mechanisms contributing to the HVR. Finally, we suggest important frontiers for future investigation. To keep the review concise, we will restrict attention to responses to isocapnic changes in Pa_{O_2} . Other factors which decrease arterial O_2 content (e.g. anemia, carbon monoxide) or ventilatory drive (e.g. decreased CO_2 drive with hyperventilation from hypoxia) will not be considered. Also, we focus on air breathing vertebrates, and primarily mammals, since they have been used for most studies reported in the literature. Interesting differences in the HVR with development (Haddad et al., 1995) or aging and gender (Tatsumi et al., 1995) are not considered here. Many of the mechanisms we consider have been discussed in more detail in other reviews (Eldridge and Millhorn, 1986; Weil, 1986; Bisgard and Neubauer, 1995; McCrimmon et al., 1995), and for conciseness these are cited in place of original references where possible.

2. Mechanisms during or following brief hypoxic exposures

Changes in ventilation (including its components \dot{V}_T and f_R) during or following a brief hypoxic exposure (2–5 min) are shown schematically in Fig. 1A,B. Although the primary stimulus to these changes is a decrease in Pa_{O_2} acting on

the peripheral (carotid body) chemoreceptors, similar responses with similar time course have been elicited in anesthetized, paralyzed mammals by electrical stimulation of the carotid sinus nerve. Within this relatively short time domain, at least three distinct mechanisms have been identified: the acute response, short term potentiation and short term depression, (Fig. 1B).

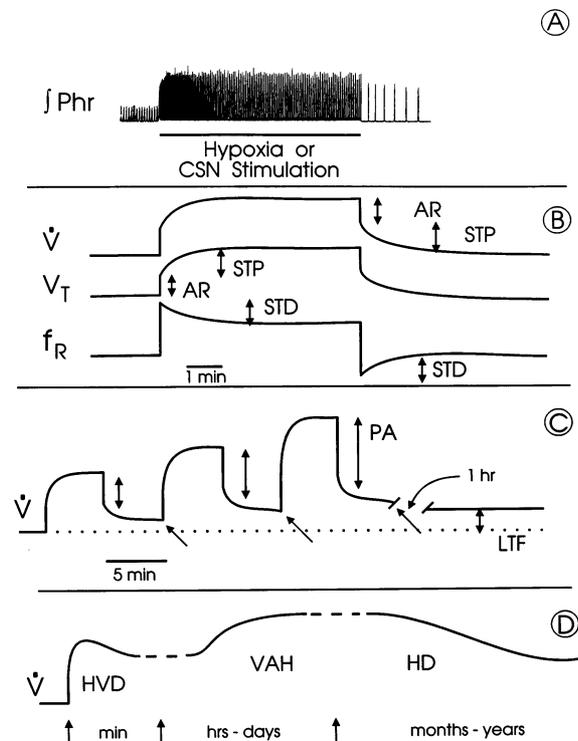


Fig. 1. (A) Phrenic nerve response to arterial chemoreceptor stimulation with hypoxia or direct electrical stimulation (bar = 120 sec) of the carotid sinus nerve (CSN) in an anesthetized rat. Frequency and amplitude increase immediately and then frequency declines during the stimulation period. Amplitude remains greater than control and frequency is depressed immediately after stimulation. (B) Ventilatory responses after brief (sec to min) hypoxic exposures include the acute response (AR), short term potentiation (STP) and short term depression (STD). (C) Ventilatory responses during and after episodic hypoxic exposures include progressive augmentation (PA) and long term facilitation (LTF). (D) Ventilatory responses during and after prolonged hypoxic exposures include hypoxic ventilatory decline (HVD), ventilatory acclimatization to hypoxia (VAH) and hypoxic desensitization (HD). Ventilatory deacclimatization to hypoxia (VDH) is not shown.

2.1. Acute response (AR)

The acute response is the immediate augmentation of ventilatory activity at the onset of hypoxia (within one breath of P_{aO_2} changing at the carotid bodies), and decrease in ventilatory activity at the termination of hypoxia (Fig. 1A). All animals studied show a hyperbolic increase in ventilation as a function of decreasing P_{aO_2} (Bisgard and Neubauer, 1995). The ventilatory response is a linear function of arterial O_2 saturation down to 70%, but this is a consequence of the shape of the O_2 -dissociation curve and the relationship between O_2 consumption and O_2 delivery at the receptor site; the adequate stimulus for carotid body chemoreceptors is P_{O_2} , not O_2 content or saturation per se.

The AR represents the effects of changes in arterial (peripheral) chemoreceptor afferent input to glutamatergic (and perhaps other types of) synapses in the nucleus of the solitary tract, NTS (McCrimmon et al., 1995). This synaptic input is 'gated', such that the immediate response to changes in afferent input is different during different phases of the ongoing respiratory cycle (Eldridge and Millhorn, 1986). Although the AR consists of changes in both respiratory timing and amplitude, the pattern of change in fR vs. V_T is highly variable and depends on species. The AR results from reflex activation of a wide array of respiratory muscles including the phrenic (Eldridge and Millhorn, 1986), accessory inspiratory (Fregosi and Mitchell, 1994), expiratory (Smith et al., 1993) and upper airway muscles (Bartlett, 1986). As defined here, the AR terminates within one breath after the afferent input (elicited by hypoxia) has returned to normal, i.e. within 10 sec.

2.2. Short term potentiation (STP)

STP represents a further, progressive increase in ventilation following AR and proceeds with a time course of many seconds up to 1 min (Eldridge and Millhorn, 1986). STP is also manifest as a slow return of ventilation to its original baseline after carotid sinus nerve stimulation, with a slightly longer time constant of 1–2 min. Al-

though STP is generally thought to be a reflection of the same mechanism during the 'on' and 'off' responses, this hypothesis has not been rigorously tested. STP is most evident in V_T or the amplitude of phrenic neural output (Fig. 1A,B), but smaller increases in fR (not shown in Fig. 1) are also observed in some experimental preparations and species (e.g. exercising humans, Fregosi, 1991). STP was formerly referred to as 'afterdischarge', based on Sherrington's concept of a prolonged neural discharge triggered by a brief stimulation (Eldridge and Millhorn, 1986).

STP has been most commonly investigated in discrete motor outputs with electrical activation of the carotid sinus nerve in cats, dogs and rats (Eldridge and Millhorn, 1986; Hayashi et al., 1993). However, STP has also been demonstrated in the ventilatory responses following brief hypoxia in human subjects while awake (Georgopoulos et al., 1990), asleep (Badr et al., 1992) and during exercise (Fregosi, 1991), and in awake goats (Engwall et al., 1991) and ducks (Milsom et al., unpublished observations). There appear to be some differences in the time course of STP between species, or within a species under different experimental conditions (i.e. it is progressively longer going from awake to anesthetized to decerebrate animals). The apparent magnitude, but not the time course, of STP changes with baseline ventilatory drive, often determined by the prevailing level of arterial P_{CO_2} (Eldridge and Millhorn, 1986).

STP does not appear specific to carotid body afferent inputs since other respiratory stimuli elicit a similar phenomenon (Eldridge and Millhorn, 1986). During and following electrical stimulation of the carotid sinus nerve, STP is evident in the output of many motor nerves including the phrenic (Eldridge and Millhorn, 1986), inspiratory intercostal (Fregosi and Mitchell, 1994) and hypoglossal (Jiang et al., 1991). It is possible to elicit STP in one motor outflow (hypoglossal) while, at the same time, another motor outflow (phrenic) is inhibited (Jiang et al., 1991). This suggests that STP arises after the divergence of neural projections to individual motor pools (e.g. at the terminal endings of pre-motor neurons or in discrete motor nuclei), or that it may be overridden by different mechanisms at discrete locations.

Several neurotransmitters and neuromodulators (e.g. serotonin, catecholamines, opiates) have been ruled out as candidates for mediating STP in mammals (Eldridge and Millhorn, 1986). The most likely explanations for STP are: (1) presynaptic calcium accumulation along premotor pathways, thus mediating enhanced transmitter release when action potentials subsequently reach the terminal (Wagner and Eldridge, 1991); or (2) release of modulatory neuropeptides, such as substance P, at key locations in the respiratory neural control system. Although neither hypothesis has been rigorously tested, there is suggestive evidence that forms of STP can occur in regions of the NTS that receive synaptic inputs from carotid chemoafferent neurons via the release of substance P (Mifflin, 1997), or in the spinal cord synaptic pathways leading from descending bulbospinal respiratory neurons to respiratory (phrenic) motoneurons (McCrimmon et al., 1997).

STP has been suggested to play a smoothing role in the control of breathing, preventing reflex activation of the respiratory system from proceeding too rapidly and, thus, imparting system stability (Eldridge and Millhorn, 1986).

2.3. Short term depression (STD)

STD is manifest as a transient overshoot in respiratory frequency at the onset of carotid chemoafferent stimulation, or a transient undershoot in frequency (fr) at the termination of chemoafferent stimulation, that lasts many seconds to a few minutes (Fig. 1A,B). To date, STD has been demonstrated only in the fr response of phrenic nerve activity in anesthetized rats during or following hypoxia or carotid sinus nerve stimulation (Hayashi et al., 1993; Bach and Mitchell, unpublished observations). Since STD affects fr, it involves the central rhythm generator and, therefore, most likely occurs in all respiratory motor outputs. Although STD is similar in some respects to hypoxic ventilatory decline (HVD, see below), important differences lead us to classify this time-dependent response as a unique mechanism or class of mechanisms at this time. The primary differences are in the ventilatory pattern (fr decreases in STD vs. VT in HVD), time course

(many seconds to a few minutes with STD vs. many minutes with HVD), and in the neurochemical mechanisms thought to be involved.

Although STD occurs in both the 'on' and 'off' response to both electrical stimulation of the carotid sinus nerve and hypoxia, it is not certain that the mechanisms underlying these responses are all the same. Coles and Dick (1996) have recently described the ventilatory 'off response' following hypoxia as the 'post-hypoxia frequency decline' (PHFD) and demonstrated that PHFD was abolished by lesions in the ventrolateral pons. This region coincides with the A5 adrenergic area, so this observation is consistent with the observation that at least part of STD is abolished following pretreatment with α_2 -adrenergic antagonists (Bach and Mitchell, unpublished observations).

The magnitude of STD can differ between the same strain of rat supplied from different vendors (Hayashi et al., 1993; Ling et al., 1997) and the magnitude of PHFD differs between sub-strains of Sprague–Dawley rats (Bach and Mitchell, unpublished observations). This suggests that both are greatly influenced by small genetic differences. The previous history of hypoxic exposure may also affect this time domain of the HVR. For example, in one series of experiments in which the magnitude of PHFD was examined at 30 min intervals in anesthetized rats, the magnitude of PHFD had declined significantly between the first and second hypoxic episodes, even though baseline phrenic nerve activity had returned to normal during the normoxic interlude (Bach and Mitchell, 1996). These results suggest that PHFD is susceptible to a degree of 'metaplasticity' (Abraham and Tate, 1997) whereby experience (with hypoxia) alters the capacity for future plasticity (i.e. magnitude of PHFD).

It is possible that STD, as we have defined it, is unique to anesthetized rats, since it is not observed in anesthetized cats following carotid sinus nerve stimulation (Eldridge and Millhorn, 1986) nor in other mammalian species investigated (e.g. goats; Turner and Mitchell, 1997). If so, the significance of STD may be limited, although it is an interesting mechanism in its own. On the other hand, STD could be a mechanism of significance in other species during, as yet, unexamined situations such as sleep or postnatal development.

3. Mechanisms identifiable during or following episodic hypoxic exposures

Two mechanisms are revealed during and following repeated episodes of hypoxia or carotid sinus nerve stimulation, respectively. These mechanisms, illustrated schematically in Fig. 1C, are progressive augmentation and long term facilitation.

3.1. Progressive augmentation (PA)

PA refers to the increase in the magnitude of the hypoxic ventilatory response seen in successive episodes of identical hypoxic stimuli (Fig. 1C). To date, PA has been demonstrated as increased V_T or peak inspiratory nerve activity in only a few experimental situations. PA was first reported to occur in the inspiratory intercostal nerve activity of anesthetized cats during successive, 2 min episodes of carotid sinus nerve stimulation, separated by 5 min intervals (Fregosi and Mitchell, 1994). In each of three successive stimulation episodes, the peak integrated inspiratory intercostal nerve activity reached a greater level, increasing from $\approx 65\%$ of maximal activity during the first stimulus episode, to $> 90\%$ of maximal activity during the third episode. PA must be distinct from STP (see above) since it occurs during stimulus episodes separated by 5 min, which greatly exceeds the time constant of STP.

PA saturates and may be absent at higher stimulus intensities. For example, less PA was observed in the phrenic relative to intercostal neurogram during the experiments described above, probably because peak integrated phrenic activity reaches levels $> 90\%$ of maximal activity during the first carotid sinus nerve stimulation episode (Fregosi and Mitchell, 1994). When cats were pretreated with a serotonin receptor antagonist, however, peak phrenic activity during the first episode was reduced to between 60 and 70% of maximal activity, and PA was now revealed during sequential stimulus episodes (Fregosi and Mitchell, 1994). These experiments also suggest that PA does not depend on the activation of methysergide sensitive serotonin receptors and, therefore, is independent of long term facilitation (see below).

PA occurs in awake goats (Turner and Mitchell, 1997) and ducks (Milsom et al., unpublished observations) during successive bouts of hypoxia. However, in neither of these latter cases has PA been investigated following administration of serotonin receptor antagonists. Thus, it is difficult to be certain that the progressively increasing HVR in successive episodes has not resulted from the development of long term facilitation.

3.2. Long term facilitation (LTF)

During protocols involving successive episodes of hypoxia or carotid sinus nerve stimulation, respiratory motor output progressively increases during the normoxic, non-stimulated intervals (Eldridge and Millhorn, 1986). Such long term facilitation (LTF) of ventilation primarily involves increases in V_T and lasts for many minutes to several hours after the final stimulus episode (Fig. 1C). LTF appears to be uniquely elicited by episodic carotid body chemoafferent stimulation, and is not observed following continuous hypoxic exposures (Dwinell et al., 1997) or following stimulation of other respiratory afferent inputs (Eldridge and Millhorn, 1986). To date, LTF has been observed to varied degrees in anesthetized and decerebrate cats (Eldridge and Millhorn, 1986; Fregosi and Mitchell, 1994), anesthetized rats (Hayashi et al., 1993; Bach and Mitchell, 1996), awake rats (Podolsky et al., unpublished observations), awake dogs (Cao et al., 1992), awake goats (Turner and Mitchell, 1997) and awake ducks (Milsom et al., unpublished observations). In contrast, LTF is not elicited in normal awake human subjects following episodic hypoxia (McEvoy et al., 1996). There appears to be substantial differences in the magnitude and duration of LTF among species and different experimental preparations. Of particular note, it appears that LTF is more difficult to elicit, and is less robust in awake versus anesthetized or decerebrate animals. Similarly, LTF is less robust in vagally intact versus vagotomized, anesthetized cats (Mateika and Fregosi, 1997), suggesting that inhibitory vagal mechanisms can obscure the expression of LTF.

There appears to be considerable heterogeneity in the LTF exhibited by different respiratory motor outputs. For example, electrical stimulation of the carotid sinus nerve in anaesthetized cats elicits a more robust LTF in the inspiratory intercostal nerve than in the phrenic nerve (Fregosi and Mitchell, 1994). Following episodic hypoxia in anesthetized Sprague–Dawley rats, LTF in the hypoglossal nerve either exceeds (Bach and Mitchell, 1996), or is considerably less than, that in the phrenic nerve, depending on which vendor provided the rats (Bach et al., unpublished observations). Thus, available data suggest there is a considerable degree of heterogeneity among different respiratory motor outputs within an animal, or in the same motor output among different experimental preparations, species or genetic lines within a species.

Serotonin is implicated in the underlying mechanism of LTF since pretreatment with serotonin receptor antagonists such as methysergide blocks LTF (Eldridge and Millhorn, 1986; Bach and Mitchell, 1996). Furthermore, LTF is impaired following administration of the tryptophan hydroxylase inhibitor, para-chlorophenylalanine, or the serotonergic neurotoxin, 5-7, dihydroxytryptamine (Eldridge and Millhorn, 1986). Although the details of involvement of serotonin in LTF are not yet clear, available information has been recently reviewed (McCrimmon et al., 1995). LTF must involve brainstem neurons because: (1) it is elicited uniquely by peripheral chemoreceptor stimulation, and afferent fibers in the carotid sinus nerve synapse predominantly in the NTS; and (2) the cell bodies of serotonergic neurons are located in brainstem raphé nuclei. Activation of carotid chemoreceptors stimulates raphé neurons (McCrimmon et al., 1995), demonstrating the necessary linkage for serotonergic involvement in LTF. These raphé neurons project to the spinal cord and cranial motor nuclei, amongst other locations, and have the potential to augment respiratory motoneuron excitability, thereby augmenting respiratory motor output (McCrimmon et al., 1995). Most studies indicate that LTF can occur without change in \dot{V}_R (Cao et al., 1992; Hayashi et al., 1993; Fregosi and Mitchell, 1994) and, thus, LTF occurs primarily via effects on

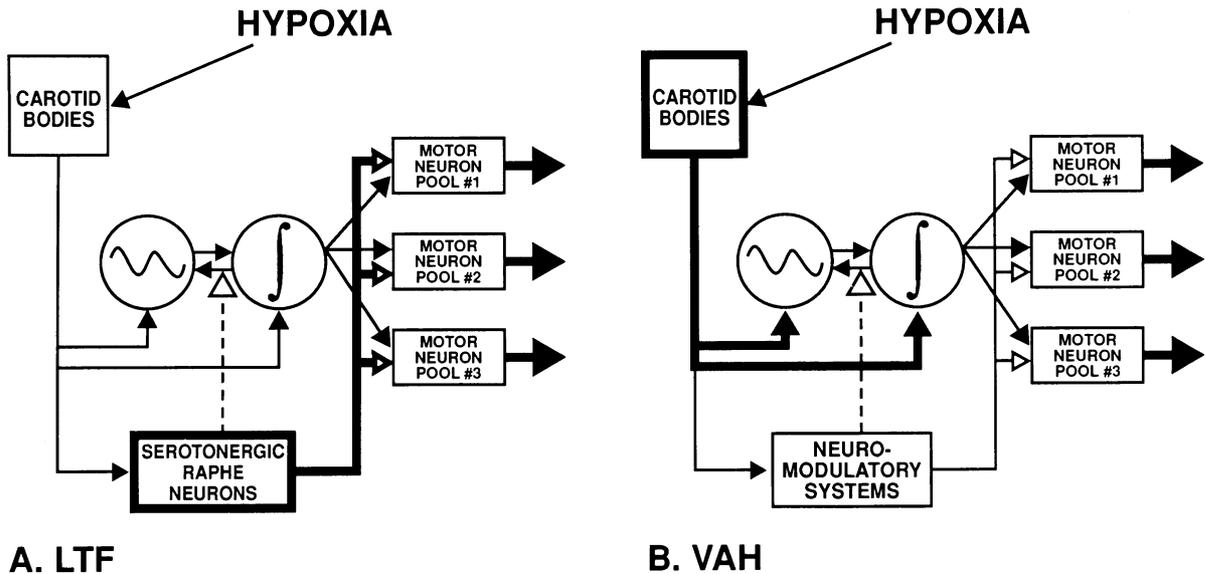
burst pattern formation versus rhythm generation. Therefore, it is possible (if not likely) that at least some of the serotonergic effects occur via actions on respiratory motoneurons. This model is schematized in Fig. 2A.

LTF may be of significance in maintaining stable breathing during sleep since episodic hypoxia and hypercapnia often result from the unstable breathing patterns that characterize this physiological state. Serotonergic raphé neurons discharge at dramatically reduced rates during sleep (Jacobs and Azmitia, 1992), particularly stage IV and REM sleep. Repeated activation of these neurons by episodic hypoxia could offset the withdrawal of serotonergic input to respiratory motoneurons, limiting or controlling the severity of respiratory muscle atonia (Kubin et al., 1992). By such control of respiratory motoneuron excitability, LTF could assure adequate ventilatory efforts and ensure upper airway patency, thereby reducing episodes of obstructive sleep apnea.

Previously, it was suggested that LTF may represent the mechanism underlying ventilatory acclimatization to hypoxia (VAH, see below) (Eldridge and Millhorn, 1986). However, as reviewed by McCrimmon et al. (1995), LTF cannot completely explain VAH. For example, rats still exhibit VAH after whole body depletion of serotonin which abolishes LTF. Potential interactions between LTF and VAH may exist but these have not been studied. For example, it is not known if LTF can be elicited by episodic exposure to deep levels of hypoxia during chronic exposure to more moderate levels of hypoxia.

4. Mechanisms identifiable during or following prolonged hypoxic exposures

Additional mechanisms become apparent when the hypoxic exposure is continuous for a prolonged period of several minutes to years. Among these mechanisms are: hypoxic ventilatory decline and ventilatory acclimatization and deacclimatization to hypoxia that occur during chronic exposures, and hypoxic desensitization that occurs with life-long hypoxia (Fig. 1D).



A. LTF

B. VAH

Fig. 2. Working models for time-dependent changes in the HVR. CNS neurons generate the respiratory rhythm and burst pattern formation of premotor respiratory neurons, which activate various motoneuron pools. Afferent input from arterial chemoreceptors can affect both rhythm and pattern formation. Neuromodulatory reflex pathways are shown with open arrows. (A) LTF occurs through increased serotonergic facilitation of respiratory motor neurons (thick lines) with no change in the O_2 sensitivity of arterial chemoreceptors (thin lines) and possibly direct effects of brainstem serotonergic neurons on medullary respiratory neurons (dashed line). (B) VAH occurs through increased O_2 sensitivity of arterial chemoreceptors (thick lines) without any apparent increase in the effectiveness of the efferent pathway (thin lines), but perhaps increased gain of the translation of afferent information into premotor neuronal output in the CNS (dashed line).

4.1. Hypoxic ventilatory decline (HVD)

HVD is the 'roll off' or decrease in ventilation relative to the acute response that occurs when moderate hypoxemia is sustained for 5–30 min in adult animals (Fig. 1D) (Bisgard and Neubauer, 1995). HVD is distinct from the secondary decrease in ventilation that arises from the hypocapnia accompanying the acute response because it also occurs during isocapnic hypoxia. As defined here, HVD also differs from the biphasic HVR observed in neonates and small rodents which represents an appropriate decrease in ventilation accompanying the decrease in metabolic rate that occurs with hypoxia, although it may share some common mechanistic features. The onset and resolution of HVD have a similar time course. Once HVD is established, the ventilatory response to subsequent hypoxic challenges is depressed for up to 60 min after normoxia is restored, but this can be shortened by breathing O_2 enriched gas mix-

tures (Neubauer et al., 1990). HVD also persists throughout chronic exposures to hypoxia, for example during sojourn to altitude (Sato et al., 1992).

HVD has been observed in awake humans, and in awake and anesthetized cats, but it could not be demonstrated in awake dogs (Bisgard and Neubauer, 1995) or rats (Aaron and Powell, 1993). It primarily affects V_T , but not rhythm generation. Hence, HVD differs from short term depression (STD, see above), which decreases f_r but not burst pattern formation and which develops over a much shorter time course. HVD has only been observed in ventilation and phrenic nerve activity, and has not been examined in other respiratory motor outputs to our knowledge.

The mechanism of HVD is controversial, but experiments on different species and preparations suggest there are specific effects of hypoxia on both: (1) the sensitivity of ventilation to O_2 ; and (2) central ventilatory drive independent of any

changes in O₂ sensitivity that occur over this time domain. Experiments from several studies of awake humans indicate that O₂ sensitivity decreases during HVD, and it has been hypothesized that HVD is a specific effect of hypoxia on arterial chemoreceptors (Robbins, 1995). Strong support for this theory comes from experiments demonstrating that during sustained hypoxia, the ventilatory response to arterial chemoreceptor stimulation by hypoxia decreases more than the response to hypercapnia. The latter was calculated using a model to partition the effects of CO₂ on arterial vs. central chemoreceptor ventilatory drives (Bascom et al., 1990).

Other studies of awake humans have found ventilatory decline during sustained hypoxia without a significant decrease in O₂ sensitivity (Sato et al., 1992). This is consistent with experiments on anesthetized animals which show that the ventilatory response to arterial chemoreceptor or carotid sinus nerve stimulation (i.e. the gain or slope of the HVR) does not decrease during several minutes of inspired hypoxia or low levels of carbon monoxide inhalation (Neubauer et al., 1990). It is also consistent with neural recordings from carotid body afferents in animals, which, with the possible exception of the rabbit, show no change in chemoreceptor activity over the time course of HVD. These data suggests that HVD is a decline in central ventilatory drive independent of changes in O₂ sensitivity of arterial chemoreceptors.

It is important to realize that a central mechanism of HVD may require arterial chemoreceptors for expression. For example, HVD could act by depressing synaptic pathways activated by arterial chemoreceptors. Therefore, the fact that carotid body denervation eliminates HVD in awake cats (Robbins, 1995) cannot be used to distinguish between these two potential mechanisms. Considering all of the above, both central (CNS) and peripheral (arterial chemoreceptor) mechanisms appear to be viable hypotheses for HVD, at least in awake preparations.

The neurochemical basis of HVD is also not clear. There is experimental evidence implicating ventilatory inhibition by adenosine, GABA, and opiates, but none of these neuromodulators can

completely explain HVD (Bisgard and Neubauer, 1995). A dopaminergic mechanism may be involved in HVD in some species since haloperidol, a D₂ dopamine receptor antagonist, eliminates ventilatory roll-off in awake and anesthetized cats (Bisgard and Neubauer, 1995), but this does not occur in awake humans (Pedersen et al., 1997). Insufficient energy substrate or changes in central chemoreceptor pH are apparently not involved in producing HVD (Neubauer et al., 1990). It is possible that the lack of agreement concerning the mechanisms which underlie HVD arises because there are actually several independent mechanisms acting in this time domain. In any case, HVD must be recognized by investigators as an important mechanism which can potentially affect measurements of the HVR (e.g. during progressive hypoxia with rebreathing). Similar to ventilatory roll-off in neonates, the biological significance of HVD is hypothesized to be energy conservation during hypoxia (Neubauer et al., 1990).

4.2. *Ventilatory acclimatization to hypoxia (VAH)*

VAH is defined as the time-dependent increase in ventilation which occurs with chronic hypoxic exposures of several hours to months (Fig. 1D) (Weil, 1986; Bisgard and Neubauer, 1995). The classic example of VAH is ventilatory acclimatization to high altitude, and its physiological significance, in terms of increasing O₂ delivery, is well known (Bouverot, 1985). The time course of VAH is species dependent, and can be 'complete' after only 4–6 h in goats or may require > 10 days in humans and rats (Bisgard and Neubauer, 1995). Most of the ventilatory change in VAH arises from increases in V_T in awake humans (Forster et al., 1974) and rats (Aaron and Powell, 1993) but from increases in both V_T and f_R in goats (Engwall and Bisgard, 1990) and cats (Tatsumi et al., 1995).

Experiments on VAH usually study the effects of chronic hypoxia and hypocapnia which occur together in healthy subjects exposed to hypoxia. Previously, it was thought that VAH resulted from changes in central chemosensitivity accompanying adjustments in CSF and arterial pH to

the chronic respiratory alkalosis. Now it is clear that this mechanism alone cannot explain VAH, although changes in brain tissue acidity induced by hypocapnia and/or hypoxia may still be involved (Bisgard and Neubauer, 1995). Recent experiments show that VAH still occurs in humans during 8 h of isocapnic hypoxia (Howard and Robbins, 1995), and in goats with vascularly isolated carotid bodies perfused by isocapnic hypoxic blood for 4–6 h (Bisgard and Neubauer, 1995).

Changes in carotid body O₂ sensitivity is now a generally accepted mechanism contributing to VAH. This is consistent with the lack of VAH in carotid body denervated animals, and the increase in the isocapnic HVR during VAH in awake animals (Bisgard and Neubauer, 1995) and humans (Howard and Robbins, 1995). In goats, carotid body chemoreceptor afferent activity increases during 6 h of hypoxia (Bisgard and Neubauer, 1995) while, in cats, carotid body O₂ sensitivity increases only after 48 h of hypoxia (Vizek et al., 1987). In both cases, this is similar to the time course required for complete VAH in each species and this increase in carotid body chemoreceptor O₂ sensitivity can completely explain VAH in goats and cats. However, it is important to understand that the 'complete' VAH that occurs after only hours does not preclude different mechanisms turning off and on during longer time domains.

One such possible mechanism is altered CNS processing of arterial chemoreceptor afferent input. Evidence for this comes from studies of awake humans and rats showing an increase in the ventilatory response to arterial chemoreceptor stimulation with doxapram after acclimatization to altitude (Forster et al., 1974; Huey and Powell, unpublished observations). If chronic hypoxia does not change the effect of this drug on chemoreceptors, then the change with acclimatization must be explained by an increase in the CNS gain of the HVR that translates chemoreceptor afferent input into ventilatory output.

Dopamine is a promising candidate for a neurochemical mechanisms underlying VAH because chronic hypoxia has large effects on tyrosine hydroxylase and dopamine synthesis, and dopamine has large effects on the carotid body (Bisgard and

Neubauer, 1995). Decreased dopaminergic inhibition at D₂ receptors in the carotid body is reported to explain VAH in cats (Tatsumi et al., 1995) but its role in goats is less certain (Bisgard and Neubauer, 1995). As discussed above, the serotonergic mechanism of LTF cannot explain VAH but the persistence or absence of LTF during acclimatization has not been studied.

Fig. 2B summarizes these results with a model of VAH. It is characterized by important changes in the O₂ sensitivity of the afferent arm of the HVR. The cellular mechanisms behind increased O₂ sensitivity during chronic hypoxia involve changes in potassium and calcium channels on carotid body glomus cells (Hempleman, 1995, 1996). Also shown in Fig. 2B is a potential increase in the CNS gain of the HVR. A mechanism in the CNS is more difficult to experimentally confirm and quantify in long time domains, such as VAH. The time required for VAH is generally too long to allow measurement of changes within a single neurophysiological preparation, and unpaired experimental designs (e.g. control vs. acclimatized groups) may require normalizing neurophysiological responses (e.g. the phrenic neurogram) while baselines are changing (e.g. normoxic ventilatory drive). Such experimental challenges explain why questions remain about mechanisms of VAH, despite it being perhaps the most studied time-dependent change in the HVR.

4.3. Ventilatory deacclimatization from hypoxia (VDH)

When normoxia is acutely restored after exposure to chronic hypoxia, ventilation and ventilatory O₂ sensitivity do not immediately return to control levels (Bisgard and Neubauer, 1995). This persistent hyperventilation in normoxia after chronic hypoxia is termed VDH, and it decays with a time course similar to the time-dependent increase in ventilation with VAH. Given the similar time courses for VAH and VDH, it was generally thought that they were the same mechanism being turned 'on' and 'off', respectively. However, recent studies have shown that VDH and VAH can be dissociated, and probably arise due to separate mechanisms. For example, VDH does

not occur after VAH in goats exposed to isocapnic hypoxia for 4–6 h, but it does occur when hypoxic exposure is accompanied by hypocapnia (Bisgard and Neubauer, 1995). This suggests that changes in CO₂-sensitive mechanisms may explain VDH. In contrast, a recent study found that hypocapnia is not necessary for VDH in humans (Howard and Robbins, 1995). The authors postulated that ‘hyperventilation-induced hyperpnea’ and potential central effects of hypoxia might explain persistent hyperventilation when normoxia is restored after chronic hypoxia.

4.4. Hypoxic desensitization (HD)

When humans experience chronic hypoxia for years or a lifetime, the HVR becomes ‘blunted’. Ventilation in hypoxia is decreased relative to normal subjects acclimatized to altitude for shorter periods of time (Fig. 1D) and ventilatory sensitivity to changes in PaO₂ is decreased (Weil, 1986). This may conserve energy, through reduced work of breathing, when other non-ventilatory modes of acclimatization (e.g. metabolic, vascular, hematological) have had time to occur (Bouverot, 1985; Weil, 1986). HD is an acquired characteristic in humans that increases with the level of altitude and time at altitude. Chinese born at sea level but living at 3658 m altitude for years show HD but Tibetan natives at the same altitude do not. However, Tibetan natives from 4400 m altitude show HD (Curran et al., 1995). This suggests that both genetic and environmental factors contribute to HD. There is disagreement about whether HD is reversible (Weil, 1986).

One of the difficulties with studying the physiological mechanisms of HD is the lack of suitable animal models. Cats show HD after 2 weeks at simulated altitudes of 5500 m, but it reverses quickly with return to normoxia, in contrast to the situation seen in humans (Weil, 1986). Reports of a blunted HVR in chronically hypoxic rats can be explained by the effects of hypocapnia and anesthesia on an HVR that is actually increased by chronic hypoxia (Aaron and Powell, 1993).

The profound changes in carotid body structure that occur with chronic hypoxia might be ex-

pected to alter chemoreceptor O₂ sensitivity, but many of these morphological changes occur before HD (Bisgard and Neubauer, 1995), so their significance to HD is not clear. Increased dopaminergic inhibition at the carotid body may be involved (Bisgard and Neubauer, 1995). Changes in both carotid body chemoreceptor O₂ sensitivity and CNS gain of the HVR are reported to contribute to HD in cats (Tatsumi et al., 1991), but these results are difficult to reconcile with those from another study which shows increased carotid body chemoreceptor O₂ sensitivity in cats after the same period of acclimatization (Barnard et al., 1987).

5. Summary and significance

5.1. Terminology

The definitions proposed here are intended to simplify communication among researchers and to promote the integration of results from different experimental preparations and laboratories. Undoubtedly, these will change as investigators make progress in understanding the different time domains of the HVR. For example, descriptors proposed today may be shown to include multiple mechanisms later, just as ventilatory acclimatization and deacclimatization to chronic hypoxia (VAH and VDH) were recognized only recently as independent mechanisms.

Given the complexity of the HVR as outlined in this review, we encourage investigators to carefully report the nature and pattern of the hypoxic stimulus used in each study. It is also important to note that the hypoxic ventilatory response includes changes in both tidal volume and respiratory frequency. The neural mechanisms modulating these two components of ventilation are generally unique, hence, we encourage investigators to report changes in both of these components as well as changes in total ventilation.

5.2. Biological significance

Presently, it is impossible to interpret the biological significance of the apparent differences

seen in the mechanisms underlying given time domains of the HVR in different species. However, diversity among species in the mechanisms producing different time domains, such as is seen with the VAH, may simply reflect convergent evolution. Natural selection may have altered different mechanisms in different lineages to produce a common physiological response that improves fitness. Mechanisms that are observed in only a few or single species, such as STD, may represent unique alterations in O₂ sensitivity to meet challenges that are unique to those species. Too little is known at present to put these apparent differences into any context.

Regardless of the causes for species differences in the time domains of the HVR, time-dependent changes must be recognized and properly controlled in comparative studies designed to demonstrate physiological adaptations to different environments. The literature is replete with studies of the HVR in animals with different types of gas exchange organ, living in different oxygen environments (from subterranean burrows to high altitude mountains) and having different oxygen demands (from near-suspended animation to maximal exercise) (Bouverot, 1985). Unfortunately, the pattern of hypoxic stimulus in most of these studies is so different (or unspecified) as to make meaningful comparisons impossible. One has to ask 'Which HVR is being compared?' to properly interpret comparative studies of the HVR.

5.3. *Clinical significance*

The different time domains of the HVR also provide an important frontier for research on respiratory disease. It is unknown which of the various time domains of the HVR occur in patients with chronic hypoxemia. Does obstructive sleep apnea induce LTF in patients? Are the normal time-dependent changes in the HVR altered in patients? Could individual differences in VAH or HVD account for differences in CO₂ retention between individuals with COPD? Determining the mechanisms of time-dependent changes in the HVR, could lead to innovative treatments for patients with chronic hypoxemia.

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