Development of the respiratory control system begins early in gestation and does not achieve "adult" form until weeks or months after birth in mammals. Until recently, maturation of respiratory control was thought of as an unfolding of a relatively fixed predetermined program. However, evidence is accumulating that respiratory control development consists of complex, dynamic interactions and is influenced by a multiplicity of factors that can alter the outcome. It is now quite clear that mammals exhibit developmental plasticity, whereby long-standing or permanent alterations in mature respiratory control are induced by experience during "critical" periods of development. The same experience occurring outside of the critical period has little or no lasting effect, indicating that the plasticity depends on time windows during ontogeny when
development can be altered in response to the external environment.

Gestation and infancy are times of tremendous vulnerability. Insults very early during embryonic development can profoundly disrupt respiratory control development in ways not apparent until later. The fetus may experience episodic or chronic hypoxia, exposure to nicotine or other toxins, and a variety of other insults with potential to alter respiratory control maturation. Premature birth profoundly alters the timing of exposure to numerous experiences, and infants of any gestational age may be exposed postnatally to episodic or chronic hypoxia, hyperoxia, and drug or toxin exposures with potential to cause disruption of neural respiratory control maturation (Fig. 1). Thus early-life experiences, rather than being regarded as “environmental noise” that may obscure the “true” genetic blueprint (147), are now seen as playing a major role in guiding normal respiratory control development as well as causing potentially maladaptive changes. Pathological conditions as well as normal phenotypic diversity in mature respiratory control may have their roots, at least in part, in developmental plasticity.

Developmental plasticity in respiratory control has received little study, although new technologies have resulted recently in a marked increase in research focused on this area. This review will describe recent advances in our understanding of mammalian neural respiratory control maturation, evidence for developmental plasticity in respiratory control, potential sites and mechanisms of “developmental plasticity” in respiratory control, and the potential for pathological developmental plasticity, with the goal of highlighting areas for further research. Although beyond the scope of the present review, developmental plasticity in re-

![Fig. 1.](image_url) The fetus and newborn may be exposed to numerous patterns of \( P_{\text{O}_2} \) variation. The effects of a given \( P_{\text{O}_2} \) level or pattern vary with age. Normal fetal arterial \( P_{\text{O}_2} \) is \(-25\) Torr in mammals, increasing in the newborn to \(-80\) Torr within hours of birth (heavy line). Common patterns of arterial \( P_{\text{O}_2} (P_{\text{O}_2}) \) variation include “hyperoxia” relative to normal fetal \( P_{\text{O}_2} (A) \), fluctuating high \( P_{\text{O}_2} \) (potentially stronger effect than steady hyperoxia) \((B)\), prenatal hypoxia \((C)\), chronic hyperoxia from birth \((D)\), chronic hypoxia from birth \((E)\), single episode of hyperoxia \((F)\), single episode of anoxia or hypoxia \((G)\), intermittent hypoxia \((H)\), adult prolonged hyperoxia \((I)\), adult prolonged hypoxia \((J)\), and adult intermittent or episodic hypoxia \((K)\). The same altered \( P_{\text{O}_2} \) exposure may have qualitatively different or even opposite effects at different ages. The first several weeks after birth is a known critical period for hyperoxia exposure (see text).
spiratory control is not unique to mammals and occurs in birds, invertebrates, and other groups (9, 72). Other reviews are available on respiratory control development and selected aspects of development-related plasticity (35, 60, 62, 96, 109, 110, 143, 150).

DEVELOPMENTAL PLASTICITY AND CRITICAL PERIODS

The term developmental plasticity has been used to describe the age dependence of various aspects of respiratory control, in other words, as a synonym for development. Others use the term to refer to age-dependent susceptibility of immature animals to environmental factors and to describe true respiratory control plasticity (see Ref. 108) occurring in immature animals. However, use of the term developmental plasticity in so many different ways has reduced its utility. In this review, plasticity in respiratory control occurring during maturation will not be considered developmental plasticity if it also occurs in the mature state and is not unique to development. The term developmental plasticity retains greater utility and meaning when restricted to long-term alterations in the structure or function of the respiratory control neural network caused by experience during pre- or postnatal formation of the respiratory control system.

The term “critical period” is closely tied to the concept of developmental plasticity. In the context of experience-dependent plasticity of respiratory control, a critical period is a time window during development devoted to structural and/or functional shaping of the neural systems subserving respiratory control. Experience during the critical period can disrupt and alter developmental trajectory, whereas the same experience before or after has little or no effect. A classical example of a critical period is altered visual cortical development caused by monocular visual deprivation only within a particular developmental time frame; monocular light deprivation in the mature animal has no effect (13). Critical periods for experience-dependent plasticity have been documented for auditory, somatosensory, and visual systems as well as other functions such as language acquisition in humans and song development in birds (13). Although critical periods are highly correlated with age, age per se may not be the sole or even main determinant of the critical period. In sensory development, experience itself is a powerful determinant of critical period duration; sensory deprivation (lack of experience) delays development of the sensory function and may prolong the critical period (13). Another key point is that critical periods for neural development may vary in a highly complex manner. In the barrel cortex of the rodent, which processes sensory information from the facial vibrissae, the durations of critical periods for receptive field plasticity vary extensively between cortical layers, ending a few days after birth in some layers and persisting for months in others (53). Although unknown at this point, it is likely that similar complexity of critical period duration exists for developmental plasticity of respiratory control, given the emerging picture of similar mechanisms across neural sensory development.

RESPIRATORY CONTROL MATURATION

Newborn mammals will not survive unless the respiratory system functions effectively at birth. This requires development of the respiratory control system in utero to a point of “readiness” such that immediately after birth the infant can generate an adequate breathing rhythm and tidal volume. However, although “ready” to function at birth, the mammalian breathing control system is immature and undergoes a prolonged period of postnatal maturation.

A relatively long gestation and postnatal development allows for prolonged periods of pre- and postnatal interaction with the environment before reaching maturity. Past concepts of respiratory control system maturation as rigidly predetermined by a genetic blueprint tended to view the role of experience as limited to triggering (e.g., chemoreceptor resetting triggered by the rise in Po2 at birth) or blocking (e.g., dopaminergic inhibition induced by hypoxia). However, the explosive increase of knowledge in neuroscience now points to a radically different view in which extremely complex interactions between genes, transcriptional factors, and other gene products shape the respiratory control system within architectural and temporal constraints, and experience plays a critical role in determining developmental outcome (8). Insights into the genetics of respiratory control development have also increased tremendously within the last decade. For example, genes such as Hox and Krox-20 are critically important for determining the future pattern of the respiratory neural network (25, 34), the RET proto-oncogene and Mash-1 gene appear to play roles in chemical breathing control maturation (22, 30), Phox2 transcription factors may determine the phenotypic plasticity of peripheral autonomic ganglion (PG) sensory neurons (20), and the homeobox gene Hoxa3 appears to be essential for carotid body development (83). Therefore, contrary to earlier views of development, we now understand respiratory neural development as the outcome of complex, timing-sensitive interactions, with numerous potential sites and critical periods for developmental plasticity both in utero and during postnatal maturation.

EXAMPLES OF DEVELOPMENTAL PLASTICITY IN RESPIRATORY CONTROL

Numerous models of developmental plasticity in respiratory control have been described, including perinatal exposure to hyperoxia, chronic hypoxia, intermittent hypoxia, hypercapnia, nicotine, stress, perinatal carotid or aortic chemoreceptor denervation, and others. Several of these have been explored in depth and are reviewed in this section, with emphasis on the concept of developmental plasticity in respiratory control, as defined above. Other potential models are discussed in the section that follows, with an emphasis on
potential sites and mechanisms. As noted above, this is an evolving field, and many areas remain unexplored.

Hyperoxia. Many years ago it was demonstrated that exposure of rats and kittens to 2 wk of 30% \( O_2 \) from birth abolished the carotid chemoreceptor response to hypoxia, although long-term effects were not studied (37, 67). Recent studies show that hypoxia exposure during a critical perinatal period permanently impairs the phrenic response to hypoxia, providing one of the clearest examples to date of developmental plasticity in respiratory control.

The first long-term study showed that adult rats exposed to a 60% \( O_2 \) environment for the first month of life exhibited markedly impaired ventilatory responses to hypoxia (95). The ventilatory response to hypoxia was not impaired when rats were exposed to hyperoxia as adults, indicating that the effect of hyperoxia was unique to perinatal exposure (95). Perinatal hyperoxia exposure did not affect the ventilatory response to \( CO_2 \). These findings indicated an extraordinary degree of developmental plasticity in the ventilatory response to hypoxia that could not be explained by nonspecific effects of hyperoxic toxicity (95).

Recently, it was shown that 1 wk of perinatal hyperoxia exposure is sufficient to blunt the adult phrenic hypoxic response (10). This finding made it possible to more precisely define the window of susceptibility to the effects of hyperoxia. The impairment of phrenic hypoxic responses was similar in 3- to 4-mo-old rats exposed to 60% \( O_2 \) during the first week, second week, and first month after birth, but hyperoxia exposure during postnatal weeks 3 or 4 had little effect (10). These findings clearly demonstrate a developmental window of susceptibility or critical period, during which a major alteration in developmental outcome can be produced by changing the external environment. Early results indicated that hypoxic responses of perinatal hyperoxia-treated rats recovered slowly over a period of months (99). However, a more recent study with a larger number of rats reported life-long impairment of hypoxic phrenic responses, even at 14–15 mo of age, after 1 mo of perinatal hyperoxia (54).

The effects of perinatal hyperoxia exposure could be due to impairment of carotid chemoreceptor \( O_2 \) sensitivity, central integration of peripheral chemoreceptor inputs, or impaired respiratory mechanics. In 2- to 5-mo-old anesthetized rats that were exposed to hyperoxia for the first month of life, the integrated phrenic nerve response (minute phrenic activity) to isocapnic hypoxia was \( \sim 50\% \) reduced compared with control rats reared in normoxia (98). Carotid denervation abolished the phrenic nerve response to hypoxia in both groups. Thus, in adult rats exposed to perinatal hyperoxia, the phrenic nerve response to acute hypoxia was also impaired, suggesting that the effect of perinatal hyperoxia was not due to altered pulmonary mechanics (98). In addition, phrenic nerve responses to carotid sinus nerve (CSN) electrical stimulation were not different in adult rats exposed to 1 mo of perinatal hyperoxia vs. rats reared in normoxia (97), suggesting that central integration of carotid chemoreceptor afferent inputs was not impaired by perinatal hyperoxia exposure. Finally, 14- to 15-mo-old rats hyperoxia exposed for the first month of life exhibited carotid body hypoplasia and no CSN response to asphyxia or intravenous NaCN injection (54). Thus multiple lines of evidence strongly support the hypothesis that carotid chemoreceptor output per se is impaired by perinatal hyperoxia exposure.

Carotid chemoreceptor \( O_2 \) sensitivity is weak in the fetus and increases during the first few weeks of life. This process is termed "resetting" (of \( O_2 \) sensitivity), and it occurs in every species studied to date (7, 24, 36, 39, 90, 105, 153). However, rather than being tightly linked to embryological or postconceptional age, carotid chemoreceptor resetting is initiated and modulated by the rise in arterial \( O_2 \) (\( P_{O_2} \)) at birth (18). Raising fetal \( P_{O_{2}} \) in utero initiates resetting early (18), whereas exposure to hypoxia from birth delays maturation of the \( O_2 \) chemoreflex response (38, 75, 139). In contrast to the effect of raising \( P_{O_{2}} \) in utero, hyperoxia exposure after birth blunts \( O_2 \) chemoreflex development (37, 67). Kittens and rats reared from birth in hypoxia or hyperoxia by birth exhibit blunted carotid chemoreceptor responses to hypoxia (37, 38, 54, 67, 145). A critical period for the effects of hyperoxia exposure on carotid chemoreceptor \( O_2 \) sensitivity has not yet been defined.

How does chronic perinatal hyperoxia impair the carotid chemoreflex response to acute hypoxia? Perinatal hyperoxia exposure induces clear morphological changes in the carotid chemoreceptor afferent pathway. Rats exposed to 60% \( O_2 \) for the first month of life showed a 41% decrease in CSN unmyelinated axons, a loss of tyrosine hydroxylase (TH) expression in PG chemoafferent cell bodies, and marked carotid body hypoplasia (42). Loss of TH expression in PG chemoafferent neurons was observed after just 1 wk of hyperoxia exposure, showing that carotid body development can be altered by increased \( O_2 \) tension within a relatively brief time. Consistent with a critical period of susceptibility, PG TH expression was not affected by hyperoxia exposure during the fourth week after birth (42).

As little as 30% \( O_2 \) for 7 days has been reported to cause carotid body hypoplasia, loss of chemoafferent axons, and loss of TH-positive chemoafferent neurons in developing rats (42). However, despite these degradations, it remains unclear whether such mild, transient, perinatal hyperoxia exposure has long-term effects. Preliminary data suggest that perinatal exposure of rats to 1 wk of 60% \( O_2 \) blunts hypoxic phrenic responses recorded at 4–5 mo of age, but 1 wk of exposure to 30% \( O_2 \) has little effect (11). If confirmed, these observations suggest that the chemoafferent degeneration and hypoplasia caused by perinatal 30% \( O_2 \) exposure is either insufficient to affect function long-term, reversible, or compensated later by other adaptations.

Survival of PG sensory neurons innervating carotid body glomus cells depends on target-derived trophic support in a developmentally regulated manner. For
example, the number of TH-expressing PG neurons decreased 73% after carotid body removal in newborn rats, whereas carotid body removal in 3-wk-old rats had no effect (74). Chronic stimulation of carotid chemoreceptor activity, by rearing rats in 12% O2 for the first week of life, caused a marked increase in the number of TH-positive neurons in the PG compared with normoxia-reared control rats (20). In addition, PG neurons in culture are supported by brain-derived neurotrophic factor (BDNF), by glial cell-derived neurotrophic factor (GDNF), and by coculture with the carotid body (40, 74), and carotid body BDNF levels are reduced by chronic hyperoxia (unpublished observation cited in Ref. 42). Thus there is strong evidence for carotid body-derived, activity-dependent support of PG chemoafferent neurons, leading to the proposal that the hyperoxia-induced derangements noted above are due to decreased target-derived trophic support. Although BDNF levels in the rat carotid body are undetectable by birth, this explanation is still plausible because PG chemoafferent neurons remain target dependent after birth (19, 74). GDNF is first detectable on embryonic day 15.5 in the mouse carotid body, and expression is still strong in newborns (40), making it a likely survival factor for chemoafferent neurons during postnatal development. In addition, ~40% of mouse PG neurons, including catecholaminergic neurons innervating the carotid body, are dependent on both BDNF and GDNF for survival (40). If GDNF expression during the early postnatal period is activity dependent, then hyperoxic suppression of carotid chemoreceptor glomus cell activity could lead to loss of postnatal GDNF support for chemoafferent neurons, resulting in blunting of hypoxic chemoreflexes.

In adult rats hyperoxia-exposed during the first month of life, the blunted hypoxic ventilatory (phrenic) response (HVR) can be partially restored by 1 wk of exposure to either sustained or intermittent hypoxia (55). This finding is somewhat unexpected because hyperoxia-induced PG chemoafferent neuron phenotypic plasticity is thought to involve alterations in neuronal survival, and an earlier study reported lifelong impairment of the HVR. Although the mechanism remains unknown, restoration of the hypoxic response by intermittent hypoxia may be via a central mechanism or possibly related to carotid body hyperplasia (55). These important findings suggest that abnormal respiratory control caused by experiences during development, such as hypoxia exposure, can be at least partially offset by respiratory plasticity later during development or in adults.

Hyperoxia-induced developmental plasticity of the ventilatory response to hypoxia could have important clinical implications. Supplemental oxygen is frequently administered to full-term and premature newborn infants in neonatal intensive care units. Premature infants with chronic lung disease who received long-term oxygen therapy exhibit markedly blunted O2 chemoreflexes that persist over time (87, 88). Premature infants with bronchopulmonary dysplasia also exhibit markedly blunted arousal responses to hypoxia during sleep (57). Thus it is critically important to understand all aspects of hyperoxia-related developmental plasticity to avoid long-term, potentially lifelong, adverse effects on breathing control.

Before birth, “normal” PaO2 in the fetus is ~23–25 Torr, which is three- to fourfold lower than normal normoxic PaO2 in the newborn after birth. Therefore, all premature infants experience “relative hyperoxia” in the sense that their PaO2 is severalfold higher than fetal PaO2 for a given postconceptional age (PCA), before the normal time of birth. This raises the question of whether premature infants may be susceptible to injury by relative hyperoxia, even when the PaO2 is within the normal range for a full-term infant. Premature infants are highly susceptible to oxidative stress, and it has been known for over half a century that O2 plays a major role in causing retinopathy of prematurity, a clear example of developmental susceptibility to the harmful effects of oxygen. In human infants, exposure to higher than normal (fetal) levels of oxygen before 36 wk PCA can cause retinal vasoobliteration and abnormal neovascularization, which can lead to retinal detachment and blindness (155). After 36 wk PCA in utero, vascularization of the retina is normally complete, and elevated levels of oxygen no longer cause retinopathy of prematurity. Thus there is a critical period or window of susceptibility to the adverse effects of hyperoxia, including relative hyperoxia.

The concept of relative hyperoxia in premature infants has received little attention with respect to breathing control. Preterm birth in lambs abolishes the normal postnatal increase of the ventilatory response to hypoxia (32), although it remains unknown whether this is due to relative hyperoxia. Essentially nothing is known about the adverse effects of mild hyperoxia, including relative hyperoxia, on breathing control development in premature infants. It is still standard practice to resuscitate newborn infants with 100% oxygen, rather than room air, although there is no evidence to support the practice and evidence for adverse effects of resuscitation with 100% O2 is growing (93, 151, 152). In addition, supplemental oxygen is widely used in the neonatal intensive care setting as well as in older infants with a variety of acute and chronic pulmonary disorders. The potential for these widespread practices to cause adverse effects on respiratory control has received little attention.

**Chronic hypoxia.** Chronic hypoxia, defined as exposure hours to weeks long, affects respiratory control differently in newborns compared with adults. In adults, chronic hypoxia increases resting minute ventilation as well as ventilatory and chemoreceptor responses to acute hypoxia (reviewed by Refs. 16, 66). In sharp contrast, in every species studied to date, chronic hypoxia in developing neonates markedly blunts ventilatory responses to acute hypoxia, largely due to decreased carotid body O2 sensitivity (38, 67, 139, 145). Thus, at different stages of development, chronic hypoxia has qualitatively opposite effects on the response to acute hypoxic challenge, suggesting the existence of...
factors involved in chemoreflex development that are unique to the developmental period.

Exposure to chronic hypoxia during postnatal maturation provides another example of developmental plasticity. Adult rats that were exposed to 6 days of hypoxia (10% O₂) during the first week of life exhibit abnormally high resting minute ventilation in room air (114) and markedly blunted ventilatory responses to acute hypoxic challenge (115). In sharp contrast, a 1-wk exposure to 10% O₂ after weaning did not affect resting ventilation or the HVR in adult rats (114, 115), indicating that the effects of chronic hypoxia were unique to a critical period or developmental window of susceptibility. Long-lasting impairment of acute hypoxic responses by exposure to chronic hypoxia during development has also been reported in species that are more mature at birth, such as cats and sheep (68, 139). Although there is no question that perinatal hypoxia causes long-term impairment of the HVR, the mechanism is still controversial. Recent preliminary data suggest that, despite impaired ventilatory responses to hypoxia while awake, phrenic neural responses to isocapnic hypoxia were not impaired in adult rats reared in 10% O₂ for the first week of life (12). These observations suggest that perinatal hypoxia may produce long-lasting impairment of the HVR, at least in rats, mainly through effects on respiratory mechanics. More studies are needed to determine the long-term effects of perinatal hypoxia on respiratory mechanics vs. O₂ chemosensitivity, central nervous system integration of respiratory signals, or chemoafferent inputs.

Gender appears to be another important influence on developmental plasticity of respiratory control. For example, compared with sea-level controls, rats reared at high altitude (3,600 m) exhibited marked long-lasting differences in onset of the ventilatory response to hypoxia during development, carotid body TH levels, and other aspects of respiratory control, some of which were dependent on the sex of the animal (82).

Developmental plasticity in carotid chemoreceptor O₂ sensitivity has also been demonstrated at the cellular level. Although mechanisms of O₂ sensing are not fully understood, the current consensus view is that hypoxia depolarizes carotid body glomus cells, leading to elevated intracellular calcium concentration ([Ca²⁺]ᵢ) via influx through voltage-gated Ca²⁺ channels, causing release of excitatory neurotransmitter(s) and excitation of apposed CSN endings. Using the [Ca²⁺]ᵢ response to hypoxic challenge as a marker of glomus cell O₂ sensitivity, studies in rats and rabbits have shown that the glomus cell O₂ sensitivity is weak in newborns and increases during postnatal development (7, 144, 145, 153). In rats reared in normoxia, glomus cell [Ca²⁺]ᵢ responses to hypoxia reach mature levels at ~2 wk of age (153). In sharp contrast, the carotid body glomus cell response to hypoxia and the postnatal increase were completely abolished by rearing rats in 12% O₂ from birth to 18 days of age (145). In rats reared from birth to 11 days in 12% O₂, the blunted glomus cell [Ca²⁺]ᵢ response to hypoxia persisted and was only partially recovered 1 wk after returning to room air. The effects of other chronic hypoxia durations, levels of hypoxia, and timing of exposure have not been examined at the glomus cell level in developing animals or in mature or adult animals; thus it remains unknown whether impaired glomus cell function is a mechanism of long-term developmental plasticity.

These findings, together with the developmental effects of hyperpoxia, indicate the existence of a “developmental window” of acceptable environmental PO₂. During a critical period of maturation, environmental PO₂ must remain between upper and lower limits or thresholds, which, when exceeded in either direction, may result in impaired development of the HVR. However, it should be emphasized that hypoxia and hyperoxia may impair the HVR by very different mechanisms.

**Transient postnatal anoxia.** A single episode of anoxia on day 2 of life has been reported to affect brain monoamine metabolism at 7–8 wk of age in rats (33), although little is known about long-lasting effects on respiratory control maturation. An early study indicated that a 10-min anoxia exposure on days 1 through 6 had no effect on respiratory control in developing rats (131). However, recently it was found that a single 20-min anoxia exposure on postnatal day 0 (day of birth) in rats resulted in no differences compared with controls at 10 days of age but an enhancement of the ventilatory response to hypoxia in the anoxia-exposed group at 25 days of age (130). If confirmed, these interesting findings constitute an example of a transient single event early in development followed by late emergence of abnormalities not detectable at an intermediate age. Essentially nothing is known about the mechanism(s) underlying this form of developmental plasticity.

**Prenatal hypoxia.** Developmental plasticity induced by prenatal chronic hypoxia exposure has received little attention. Chronic prenatal hypoxia in rats leads to respiratory and metabolic disturbances from day 1 of life (59) and neurochemical alterations in the respiratory regions of the medulla (156). Rats born after exposure to environmental hypoxia between embryonic days 5 and 20 exhibited marked augmentation of the ventilatory response to hypoxia and absent hypoxic hypometabolism at 3 wk of age, but by 9 wk of age hypoxia responses and hypoxic hypometabolism had normalized (120). Carotid body dopamine content was decreased by prenatal hypoxia and remained so at 9 wk postnatal age (120). The same gestational hypoxia exposure (10% O₂ from embryonic day 5 to 20) resulted in adrenocortical and autonomic nervous system abnormalities that persisted into adulthood (104, 119). Except for these few studies, very little is known about developmental plasticity of adult respiratory control due to prenatal hypoxia.

**Hypercapnia.** Although hypercapnia is a relatively common clinical condition during infancy, developmental plasticity in respiratory control due to perinatal...
hypercapnia has received little attention. Preliminary results from an early study in rats suggested that exposure to 7% CO₂ for the first week of life blunted the ventilatory response to acute hypercapnia at 45–50 days of age (126). A later report from the same investigators confirmed these effects 2 days after return to room air (e.g., 9 days of age) but did not report long-lasting effects in adults (125). Although nonmammalian breathing control is beyond the scope of this review, it is noteworthy that gender-dependent developmental plasticity in the avian ventilatory CO₂ response has been reported (9).

POTENTIAL SITES AND MECHANISMS OF DEVELOPMENTAL PLASTICITY IN RESPIRATORY CONTROL

Although the field of developmental plasticity in respiratory control remains relatively unexplored, recent advances in neuroscience research point to the existence of numerous potential sites where developmental plasticity appears likely. Several examples are reviewed below with the aim of pointing to avenues for developmental plasticity research in respiratory control.

Rhythmic activity in spinal motoneurons innervating the respiratory muscles is produced by a neural network located in the ventrolateral region of the brain stem. This network consists of three interconnected groups of neurons: the ventral respiratory group (VRG) in the ventrolateral medulla, the dorsal respiratory group (DRG) in the nucleus tractus solitarius (NTS) of the medulla, and the pontine respiratory group (PRG) in the dorsolateral pons. In addition, a group of neurons in the pre-Bötzing complex (PBC), within the VRG, is essential for respiratory rhythm generation (76, 85, 127). The respiratory rhythm generated by this network is not simply repetitive inspiratory bursts but consists of alternating bursts of neural activity controlling separate inspiratory, postinspiratory, and expiratory phases of the respiratory cycle (127). Generation of resting respiratory rhythm and ventilatory responses to stresses such as exercise and hypoxia depend on inputs from numerous central and peripheral sites, including central chemosensitive neurons, peripheral chemoreceptors, laryngeal and pulmonary receptors, and a variety of others (reviewed in Refs. 15, 56, 124).

Neural network development. Prenatal formation of the respiratory neural network offers high potential for developmental plasticity because a transient phase of early hindbrain segmentation profoundly affects patterning of the network and, therefore, later respiratory controller function (27). Early during embryonic development in vertebrates, the hindbrain becomes segmented into cellular compartments called rhombomeres, which form patterns that play a critical role in determining later maturation and organization of these neurons into the VRG, DRG, PRG, and PBC that make up the respiratory pattern generator. After this critical phase of development, a remarkable reconfiguration of brain stem neurons and synapses occurs (79). Therefore, early hindbrain segmentation offers a “critical window” during which the later function and organization of neurons from each rhombomere are determined. The segmentation process is controlled, at least in part, by several genes including Kriesler, Krox-20, and Hoxa-1 (79), and recent studies using mutant mice strongly suggest that insults affecting these genes can profoundly affect respiratory control maturation.

Homeobox genes regulate gene expression and control morphogenesis by determining alignment, patterning, and cell differentiation (106). Hindbrain rhombomeres 3 and 5 are eliminated by inactivation of the homeobox regulator gene Krox-20 (134); Krox-20 mutant homozygous mice exhibit a slower respiratory rate and apneas 10 times longer than those in wild-type animals at birth (79). Early expression of another gene important in hindbrain segmentation appears to be vital for breathing control after birth. Hoxa1(−/−) mice, in which rhombomeres 4 and 5 are eliminated, exhibit severely abnormal respiratory control within 24 h of birth (27). Another segmentation gene, kriesler, affects rhombomeres 5 and 6, and its deletion (−/−) results in a nonlethal decrease in tidal volume and frequency in newborn mice (27). On the basis of comparisons of mice with various targeted deletions, it was suggested that perturbations in developmental trajectories determined by early hindbrain segmentation may disrupt development of a pontine antiapneic system and cause abnormal chemosensory control of breathing (26). Thus modification of hindbrain segmentation profoundly alters maturation of the respiratory neural network. Such findings suggest the existence of a critical period during hindbrain segmentation, during which perturbation of rhombomeres 3 or 5 can alter development of a pontine respiratory rhythm-promoting function, leading to severe respiratory control abnormalities after birth.

Other homeobox genes have been reported to cause developmental respiratory control abnormalities in immature mice with targeted deletions. Targeted deletion of the POU domain transcription factor Tst-1/Oct-6/SCIP, which is expressed transiently during myelination, leads to a severe defect in peripheral myelination (14). Tst-1/Oct-6/SCIP is expressed in the NTS, and in Tst-1/Oct-6/SCIP-mutant homozygous mice, aberrant differentiation and migration of specific neurons are associated with severe breathing defects in newborns (14); it has been suggested that neurons in the NTS require Tst-1/Oct-6/SCIP for development of a normal respiratory phenotype (58). The Rnx gene has also been shown to play a role in proper formation of the NTS, the target for visceral sensory afferents, and is expressed in the developing dorsal and ventral regions of the medulla oblongata. Rnx-deficient mice exhibit central respiratory failure after birth, suggesting that Rnx is important for the development of the ventral medullary respiratory centers (122, 137).

Genes involved in neural crest development may also be important sites for developmental plasticity. RET
protooncogene expression plays a key role in neural crest development and is expressed in the mouse hindbrain (58). Interestingly, RET-null mutant newborn mice exhibit abnormal breathing at birth and have been shown to have depressed ventilatory responses to CO₂ (22); RET gene mutations have also been reported in children with congenital central hypoventilation (CCHS) (2, 132, 133). Mutant mice deficient for the Mash-1 gene, also important in neural crest development, appear to have impaired ventilatory responses to hypoxia and hypercapnia, suggesting a role in maturation of chemical breathing control (31). Recent evidence from the same group (30) indicates that Mash-1 is upstream of RET expression in noradrenergic brain stem neurons important for respiratory rhythm modulation, suggesting that the phenotype of Mash-1 mutants may be due to impaired RET gene expression.

Development of the central respiratory rhythm also requires BDNF, which is one of the neurotrophin classes of growth factors (41). Newborn transgenic mice lacking BDNF have severely irregular and depressed breathing as well as a reduced chemosensory drive (41). Brain stem-spinal cord preparations from BDNF(−/−) mice exhibit a markedly reduced central respiratory discharge frequency, indicating that BDNF is required for development of normal respiratory rhythm generation (4). This effect was evident on postnatal day 1, indicating that BDNF is required for normal central respiratory network development before birth. BDNF was not required for development of ventilatory CO₂ sensitivity in mice (41). The potential for BDNF-related plasticity in respiratory control is unknown because little is known about early-life experiences that affect BDNF-dependent development. However, it was recently demonstrated that early maternal deprivation in rats, on postnatal day 9, significantly reduced BDNF expression in the hippocampus during adulthood (129). Interestingly, reduced hippocampal BDNF expression after early maternal deprivation was not immediately present or present in preweaning animals (129).

Primary sensory neuron plasticity. BDNF is crucial for normal development of peripheral chemosensory drive as well as central respiratory output. Two-day-old wild-type mice exhibit an ~30% drop in minute ventilation when exposed transiently to 100% O₂, indicating substantial peripheral chemosensory drive. In sharp contrast, BDNF-deficient newborn mice (−/−) showed no change in minute ventilation with 100% O₂, indicating absence of oxygen-mediated drive under resting conditions (41). BDNF is also required for arterial baroreceptor system development; baroreceptor regions in BDNF(−/−) mice are completely devoid of innervation (19).

Potential for a critical period exists with respect to the timing of BDNF mRNA and protein expression. BDNF expression for both the arterial baroreceptors and chemoreceptors coincides with onset of sensory innervation, which is approximately embryonic day 13.5–16.5 in mice, consistent with its role as a target-derived survival factor during innervation (19). A recent study found that both BDNF and GDNF are required in mice, between embryonic days 15.5 and 17.5, for survival of neurons, including dopaminergic neurons, in the nodose-PG complex (40). GDNF(−/−) mice deficient in PG neurons exhibit abnormal breathing after birth, characterized by reduced respiratory frequency and increased episodes of apnea (40).

GDNF dysfunction may play a role in CCHS. These infants have an abnormally low chemical respiratory drive and irregular breathing manifested in hypventilation and hypoxemia after birth. Mutations of both RET and GDNF genes have been reported in cases of CCHS (2, 84, 132, 133); in addition, as noted above, RET-deficient newborn mice exhibited depressed ventilatory responses to hypercapnia (22). Therefore, although much more research is needed to determine factors that may influence maturation, it is clear that development of the chemosensory pathway presents numerous possibilities for developmental plasticity in respiratory control.

Another potential mechanism of developmental plasticity is activity dependence of transmitter phenotype in PG sensory neurons that innervate the oxygen-sensing cells of the carotid body. Under normal conditions, only a small number of PG sensory neurons, ~10–20%, exhibit dopaminergic traits such as TH expression (51, 86). However, when PG neurons in culture were stimulated with depolarizing concentrations of KCl, TH expression was induced in 100% of PG sensory neurons (73). The activity dependence of TH expression in PG sensory neurons is developmentally regulated. Depolarization induced TH expression in 92% of PG sensory neurons on embryonic day 16.5 but was fivefold less effective by postnatal day 7 (21). In addition, in dissociated embryonic day 16.5 PG neurons, a brief transient period of depolarization led to a long-term potentiation of TH expression in response to subsequent depolarizing stimuli (21). Together, these findings suggest that PG sensory neuron dopaminergic phenotype in vivo depends on electrical activity during a critical period of sensitivity to the effects of depolarization. This is consistent with the hypothesis that early tonic activity of carotid body afferents determines transmitter phenotype of PG sensory neurons in vivo (21). Activity dependence of PG sensory neuron transmitter phenotype, during a critical period of development, presents a high potential for developmental plasticity in primary and possibly secondary sensory neurons. BDNF release from PG sensory neurons has also been shown to be activity dependent (5). As discussed above, hyperoxic suppression of carotid body activity during a critical period has been proposed as a mechanism by which perinatal hyperoxia could impair oxygen chemosensory maturation.

Maturation of the HVR. Although knowledge of HVR maturation has increased tremendously during the past decade, development of the underlying neuroanatomic and neurophysiological substrate is still quite poorly delineated. Available data present a picture of remarkable complexity involving activity-dependent development of chemosensory pathways and brain structure; however, the specific actions of neurotrophic factors during this process are still poorly understood. The effect of single daily injections of BDNF on respiratory control is illustrated in Figure 5. BDNF is required for development of normal respiratory discharge frequency, indicating that BDNF is required for normal central respiratory network development (22). BDNF is also required for arterial baroreceptor system development; baroreceptor regions in BDNF(−/−) mice are completely devoid of innervation (19).
stem respiratory regions, as well as multiple membrane receptors, neurotransmitters, and signaling pathways, many of which develop mainly during the postnatal period (60). As noted in Examples of Developmental Plasticity in Respiratory Control, peripheral chemoafferent pathways can be profoundly deranged by hypoxia or hyperoxia during development, underscoring the potential for impairment of activity-dependent development in brain stem areas processing these inputs, such as the NTS (157). Multiple sites of activity-dependent plasticity during development raise the possibility that repetitive insults in an infant (e.g., repetitive hypoxia due to apnea) could alter the trajectory of respiratory control maturation. For example, intermittent hypoxia attenuates the HVR in developing piglets (154), impairs the autoresuscitation response (gasping) in developing rats (61), and impairs hypoxic arousal and ventilatory, upper airway, and blood pressure responses during active sleep in lambs (45, 46, 50, 80, 81). Thus the high potential for “maladaptive respiratory plasticity” in the HVR makes this a critically important area for further research. Potential neural sites for developmental plasticity in the HVR were recently reviewed (60).

Neuronal apoptotic response to hypoxia. It has been known for over 100 years that newborn mammals are more hypoxia tolerant than adults (reviewed in Ref. 138) and that neurons in developing mammals become less hypoxia tolerant with age (64). One way that hypoxia can affect developing mammals is through effects on apoptotic cell death, which is a normal component of postnatal development of the nervous system, including neurons involved in respiratory control. Rats studied at 2, 5, 10, 15, 25, 20, 60, and 120 days of age exhibited a remarkable age-related susceptibility to intermittent hypoxia exposure (10% O₂ every 90 s for 48 h) (63). Intermittent hypoxia increased apoptosis in the hippocampus and cortex at all ages. The hypoxia-induced increase in apoptosis was least at 2–5 days, peaked between 10 and 25 days, and then declined to intermediate levels between 30 and 120 days (63). These findings suggest that the outcome of intermittent hypoxia exposure is likely to be age dependent; that is, that the time frame of marked apoptotic vulnerability to hypoxia may constitute a critical period. At this time, very little is known about apoptosis in respiratory control pathways and virtually nothing is known about the developmental plasticity that may result from this mechanism (reviewed in Ref. 62).

N-methyl-D-aspartate-receptor activity. N-Methyl-D-aspartate (NMDA) receptors play an important role in respiratory control, and their maturation presents another opportunity for developmental plasticity. Therefore, it is of interest that mice lacking the NMDA receptor 1 subunit (NR1) exhibit severe abnormalities of respiratory control development. NR1(–/–) mice, studied within hours after birth, exhibited marked depression of the respiratory rhythm and overexpression of synaptic long-term depression in the NTS in vitro (121). More important for potential developmental plasticity, these abnormalities were not observed in wild-type newborn mice after pharmacological blockade of NMDA receptors, indicating that NMDA receptors are important for prenatal respiratory control development. The observation that mice do not show NMDA receptor binding during midgestation suggests the existence of a critical period, during late fetal maturation, for normal development of respiratory rhythm and synaptic transmission (121). In addition to a prenatal critical period of development, the normal postnatal increase of NMDA expression in the NTS (113) may be vulnerable to insults during critical times. For example, nicotine exposure in maturing rats between postnatal days 8 and 12, but not earlier or after postnatal day 19, disrupts development of glutamate synapses in the auditory cortex (3). Thus, depending on the type of insult and site involved, there appear to be multiple critical or susceptible periods during which NMDA receptor development can be altered.

PBC development. The PBC is considered to be a major center of respiratory rhythmogenesis (56, 123, 142) and therefore is a potentially important site for developmental plasticity in respiratory control. An initial study of PBC maturation found an increase in cytochrome oxidase activity (as a marker of neuronal functional activity) in the PBC of developing rats between 0 and 21 days of age and a transient decrease or plateau in cytochrome oxidase activity at postnatal days 3–4, suggesting a period of developmental reorganization or synaptic adjustment (101). The same investigators later reported two distinct critical periods of PBC postnatal development in rats (postnatal days 3–4 and postnatal day 12) when cytochrome oxidase activity and immunoreactivity for NR1 decline. They speculate that these constitute “vulnerable periods” during which inhibitory drives predominate, rendering an infant more susceptible to exogenous insults or stress (100). Although these findings are intriguing, it remains to be determined whether they are associated with functional deficits in respiratory defensive responses to hypoxia, hypercapnia, or other stressors.

Carotid body formation. Formation of the third pharyngeal arch, where the carotid body rudiment forms (92), depends on the homeobox gene Hoxa3 (28). Although little is known about mammalian carotid body development, available data suggest that the carotid body rudiment contains neural cells from the superior cervical ganglion and neural crest-derived mesenchymal cells that differentiate into glomus cells when in contact with glossopharyngeal and sympathetic nerve fibers. Hoxa3(–/–)-mutant mice have no carotid bodies at all and die at birth, although the superior cervical ganglion and CSN appear to develop normally (83). Indeed, the superior cervical ganglion in homozygous Hoxa3-mutant mice is hypertrophied, possibly as a result of lacking its target organ. Thus the embryological period of third pharyngeal arch/pouch formation is crucial for establishment of the carotid body primordium, and differentiation of neural crest mesenchymal cells into glomus cells appears to depend on innervation by the CSN, offering several potential sites and
critical periods for developmental plasticity. Virtually nothing is known about modulators and potential modifiers of these crucial developmental phases in carotid chemoreceptor formation.

**Carotid body denervation.** Surgical denervation of the carotid bodies in newborn rats leads to severe abnormalities of respiratory control and increased mortality (77, 78, 135). On the other hand, carotid denervation in more mature species such as goats or piglets does not lead to significant mortality, and other effects are mild and transient (102, 103). Long-term recovery and plasticity after denervation will be discussed in another article in this series (52) and is beyond the scope of this review. However, studies that use carotid body denervation (CBD) point to a postnatal period of significant “developmental vulnerability.” Rats denervated at 7–8 days of age exhibit impaired ventilatory responses, decreased weight gain, and developmental delay, whereas those denervated before or after showed milder or minimal effects (135). In addition, rats in the same study exhibited a significant ventilatory response to hypercapnia at 1–3 and 12–14 days of age but not on postnatal days 6–7. Interestingly, another study of carbon dioxide ventilatory response maturation in rats reported a nadir in carbon dioxide chemosensitivity on postnatal day 8 (146). Age-dependent vulnerability to the effects of CBD was also reported for piglets, showing profound oxygen desaturation, prolonged apnea, irregular breathing, and tachycardia when carotid denervated at 12–15 days but not before or after (29). A recent study of CBD in piglets, using a lateral surgical approach, did not find such severe abnormalities but did report post-CBD hypoventilation after 10 days of age but not before (136). Taken together, these studies strongly suggest the existence of postnatal, age-dependent windows of susceptibility during which lack of carotid chemoreceptor input results in significant vulnerability to stressors such as hypoxia.

**Nicotine exposure.** Developmental neurotoxicity from nicotine exposure is well documented (43). Prenatal nicotine exposure during the last third of gestation alters postnatal breathing pattern in lambs up to 3 wk of age, although it remains unclear whether the mechanism involves altered lung mechanics, respiratory control, or both (65). Mice exposed prenatally to nicotine exhibited apnea after hypoxia and altered hypoglossal inspiratory neuron output during the early postnatal period (128). Autoresuscitation, a critically important defensive response in newborns, is impaired in rat pups during the first week of life by prenatal nicotine exposure (47, 48). Nicotine-related developmental plasticity in human respiratory control has received relatively little attention to date, partially because of the large sample sizes needed to detect small effects on responses to mild challenge (148). One study found that human infants of mothers who smoked during pregnancy showed impaired arousal responses during sleep in response to hypoxia challenge but not carbon dioxide challenge (94). In another study, no differences in the response to asphyxial challenge were demonstrated between maternal smoking-exposed infants vs. nonexposed infants, although there was a trend showing more ineffective responses in the smoke-exposed group (23). Together, it is clear that prenatal nicotine exposure impairs several aspects of respiratory control during postnatal development. Whether pre- or perinatal nicotine exposure produces alterations in respiratory control in adults remains unknown.

**Sudden infant death syndrome.** Sudden infant death syndrome (SIDS), the leading cause of mortality in infants between 1 wk and 1 yr of age, is defined as the sudden death of an infant, unexpected by clinical history, that remains unexplained after a thorough postmortem examination and death scene investigation (158). Although a discussion of potential mechanisms is beyond the scope of this review, several features of SIDS deserve comment in the context of developmental plasticity of respiratory control. Age at death in SIDS presents a striking pattern, showing a peak between 2–4 mo of age, with the great majority of SIDS deaths occurring before 6 mo. This finding alone strongly suggests a developmental “window of vulnerability” related to the physiological cause(s) of death. Numerous lines of evidence suggest that some infants who succumb to SIDS exhibit abnormalities in brain stem regions involved in cardiorespiratory control (91, 112, 117, 118) as well as physiological signs of profound hypotension and inadequate compensatory responses (70, 71).

Although the causes of brain abnormalities, autonomic dysfunction, and impaired cardiorespiratory compensatory responses in SIDS infants remain unknown, the emerging picture of developmental respiratory control plasticity suggests that functional and even structural abnormalities in cardiorespiratory control pathways may result from experience (environmental interactions) during development. Pre- and postnatal smoke exposure is strongly linked in a dose-dependent manner to increased SIDS risk (17, 107, 116, 159), prenatal smoke exposure alters development of nicotinic binding in three brain stem nuclei related to arousal or cardiorespiratory control (111), and animal studies link pre- and perinatal nicotine exposure with impaired cardiorespiratory defensive mechanisms (6, 44, 48, 49, 65, 140, 141, 148). SIDS is also strongly linked with the prone sleeping position; the incidence has recently been cut in half or more by public health campaigns that promote the supine sleeping position for infants (1). The mechanism relating SIDS and sleeping position remains unknown but probably involves positional effects on breathing as well as effects of sleep position on arousal and autonomic cardiorespiratory compensatory responses (28a, 70). In addition, SIDS risk is increased by bedding materials that increase the probability of rebreathing hypercarbic/hypoxic exhaled gases (89, 149), suggesting that infants sleeping prone may experience intermittent hypoxia or asphyxia, known to alter development of the HVR as discussed in *Maturation of the HVR*. This raises the potential for maladaptive devel-
opmental plasticity in cardiorespiratory defensive and compensatory mechanisms, possibly associated with repetitive hypoxic episodes during a vulnerable period of development.

SUMMARY

Far from being predetermined by a relatively fixed genetic blueprint, respiratory control system development is strongly influenced by experience. The role of experience clearly goes far beyond simple triggering and blocking; experience during pre- or postnatal periods (e.g., hypoxia) can profoundly alter respiratory control system development in complex ways. Although some examples of clear-cut developmental plasticity have been identified (e.g., perinatal hypoxia) and numerous potential sites and mechanisms have been described, the full extent and potential importance of developmental plasticity in respiratory control remains largely unknown. Recent studies of respiratory control development have begun to identify critical genes, gene products, and periods of development, but little is known about the environmental perturbations that potentially influence these factors. In addition, although a great deal of progress has been made by studying animal models, very little is known about developmental plasticity in human respiratory control and the role of pre-, peri-, or postnatal experience in normal phenotypic diversity or the pathogenesis of human disease.

REFERENCES


oxia in maturing piglets is unrelated to NTS ME or SP levels.