Respiratory Physiology & Neurobiology 185 (2013) 67–74

Contents lists available at SciVerse ScienceDirect

Respiratory Physiology & Neurobiology

journal homepage: www.elsevier.com/locate/resphysiol

Review

Voltage-gated Na^+ channels in chemoreceptor afferent neurons—Potential roles and changes with development☆

David F. Donnelly*

Department of Pediatrics, Yale University School of Medicine, 333 Cedar St., New Haven, CT 06510, USA

ARTICLE INFO

Article history:
Accepted 10 August 2012

Keywords:
Carotid body
Na^+ channel
Post-natal maturation
Action potential

ABSTRACT

Carotid body chemoreceptors increase their action potential (AP) activity in response to a decrease in arterial oxygen tension and this response increases in the post-natal period. The initial transduction site is likely the glomus cell which responds to hypoxia with an increase in intracellular calcium and secretion of multiple neurotransmitters. Translation of this secretion to AP spiking levels is determined by the excitability of the afferent nerve terminals that is largely determined by the voltage-dependence of activation of Na^+ channels. In this review, we examine the biophysical characteristics of Na^+ channels present at the soma of chemoreceptor afferent neurons with the assumption that similar channels are present at nerve terminals. The voltage dependence of this current is consistent with a single Na^+ channel isoform with activation around the resting potential and with about 60–70% of channels in the inactive state around the resting potential. Channel openings, due to transitions from inactive/open or closed/open states, may serve to amplify external depolarizing events or generate, by themselves, APs. Over the first two post-natal weeks, the Na^+ channel activation voltage shifts to more negative potentials, thus enhancing the amplifying action of Na^+ channels on depolarization events and increasing membrane noise generated by channel transitions. This may be a significant contributor to maturation of chemoreceptor activity in the post-natal period.

1. Introduction.

Carotid body chemoreceptors transduce a decrease in arterial oxygen tension or decrease in pHa into increased action potential (AP) activity on the sinus nerve. This increased activity is essential in mediating the increased drive to breathe during hypoxia and also plays important roles in arousal from sleep and increasing sympathetic activity. The sensitivity to hypoxia undergoes a significant developmental change being relatively insensitive at birth and increasing to adult levels over the first weeks to month of life (Kholwadwala and Donnelly, 1992; Marchal et al., 1992; Mulligan, 1991; Pepper et al., 1995). This is likely due to the changing definition of 'hypoxia' at birth since the normal fetal PaO_2 (i.e., 'normoxia') is about 30 Torr and rises to 90 after birth. This developmental increase is also dependent on the oxygen level in the post-natal period. Birth into an hypoxia environment appears to ablate the signal to evoke resetting of chemoreceptor sensitivity and results in a greatly reduced ventilatory response to acute hypoxia (Eden and Hanson, 1987; Hanson et al., 1989). Perhaps surprisingly, birth into an enriched oxygen atmosphere also results in a reduction in the hypoxic ventilatory response (Eden and Hanson, 1986).

An understanding of these developmental factors is dependent on understanding the mechanism by which hypoxia results in increased nerve activity. Here, the glomus cell, a secretory cell within the carotid body and associated with chemoreceptor nerve endings, is believed to play a central role. At least some glomus cells respond to acute hypoxia with a depolarization, thereby activating voltage-dependent calcium channels and allowing an influx of calcium (Buckler and Vaughan-Jones, 1994; Wasicko et al., 1999, 2006). The rise of intracellular calcium concentration triggers secretion of dense-cored vesicles and, perhaps, cleared cored vesicles, which are often present in glomus cells (McDonald and Mitchell, 1975). The vesicular release likely contains one or more excitatory neurotransmitters that initiate action potentials in the afferent nerve endings, but we have not yet achieved a clear identification of the excitatory agents (Reyes et al., 2007b).

This last step – the initiation of action potentials – is the primary focus of this brief review, and, in particular, the potential role or roles for voltage-activated Na^+ channels in this process. The normal assigned role of Na^+ channels is supporting action potential propagation in which a depolarized portion of an axon will provide a current sink that depolarizes a neighboring portion of the nerve

☆ This paper is part of a special issue entitled "Development of the Carotid Body", guest-edited by John L. Carroll, David F. Donnelly and Aida Bairam.

* Tel.: +1 203 785 4999; fax: +1 203 785 6337.
E-mail address: David.Donnelly@Yale.edu

1569-9048/$ – see front matter © 2012 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.resp.2012.08.009

fiber. However, Na^+ channels may also play an important role in amplifying depolarizing events secondary to the release of neuromodulators or neurotransmitters, and thus dictate the relationship between transmitter release and initiation of APs.

2. Sodium channels in synaptic transmission

2.1. Importance of sodium channels in post-synaptic AP initiation

Transmission at many synapses is relatively independent of the type or number of Na^+ channels. For instance, the neuromuscular junction transmits APs over the synapse in a 1:1 fashion with a safety factor estimated at 5–7 times (Wood and Slater, 2001). That is, the magnitude of the post-synaptic depolarization caused by a presynaptic AP is 5–7 times greater than required to transmit across the junction (Wood and Slater, 2001). Thus, a loss of Na^+ channels or Na^+ channel current due to application of drugs or a reduction in extracellular Na^+ concentration has little effect on neuromuscular transmission at low blocking levels.

Most synapses, however, do not demonstrate a 1:1 coupling but rely on the summation of multiple excitatory post-synaptic potentials (EPSPs) from one or more sources to generate an AP. In this case, factors that stabilize or hyperpolarize the postsynaptic membrane would serve to inhibit AP initiation and factors which enhance EPSP magnitude would facilitate AP generation. One such factor is synaptic strengthening by modification of pre-synaptic and post-synaptic sites (Davis and Goodman, 1998). In addition, EPSPs can also be enhanced by voltage-dependent activation of Na^+ channels which have a threshold for activation around the resting potential (Crill, 1996). This has been shown to boost the magnitude of the EPSP and result in enhanced AP generation in multiple systems (Crill, 1996; Schwindt and Crill, 1995; Stuart and Sakmann, 1995).

2.2. Na^+ channel isoforms

Based on the pioneering work of Hodgkin and Huxley using squid giant axon, the activation and inactivation of Na^+ channels can be modeled using an activation scheme of the binding of three activation particles and the binding of a single inactivation particle (Fig. 1) (Hodgkin and Huxley, 1952). In the original work, Na^+ currents of clamped squid axons were elicited using voltage protocols similar to that used in Fig. 2. An ensemble average of Na^+ current was measured as a function of voltage and time and fit to power function of exponentials. In the model, the gates for activation (m) and inactivation (h) are controlled by a membrane-bound charged particles whose position (bound or unbound) is controlled by an energy barrier. As the membrane is depolarized, the energy barrier is lower and the probability for binding to the gate is higher.

This model has proven accurate in characterizing the activation and inactivation characteristics of nine isoforms of voltage gated Na^+ channels found in humans, termed Nav1.1-Nav1.9. These channels differ in their voltage of activation and voltage of inactivation as well as tissue localization. For instance, Nav1.2 is the primarily isoform in the central nervous system and in unmyelinated axons (Boiko et al., 2003; Yao et al., 2002); Nav1.8 and Nav1.9 are predominantly found in small (pain) neurons of the peripheral nervous system (Akopian et al., 1999; Djouhri et al., 2003); Nav1.7 significantly mediates pain sensing (Chahine et al., 2005; Nassar et al., 2004). A more complete description of tissue localization is given elsewhere (Chahine et al., 2005). In broad terms, isoforms can be grouped into tetrodotoxin sensitive (TTX-S) subtypes (Nav1.1, 1.2, 1.3, 1.4, 1.6, 1.7) and (relatively) TTX-resistant (TTX-R) (Nav1.5, 1.8, 1.9) (Chahine et al., 2005) based on the ability of the toxin to block the channel. The voltages of activation and inactivation for TTX-R isoforms are about 10 mV more positive than TTX-S isoforms and, thus, are less likely to participate in the initiation of AP initiation, since the activation voltage is farther removed from the resting potential.

2.3. Kinetics of activation and inactivation of Na^+ channels

Besides increasing the magnitude of depolarization events caused by external events (e.g. EPSPs), the voltage-dependent activation of an inward (i.e., depolarizing) current around the resting potential can lead to instability of the membrane potential, characterized as spontaneous oscillations. Nature utilizes these oscillations in a number of neuronal networks. For instance, rhythmic oscillations due to sodium currents generated around the resting potential underlies the rhythmic discharge behavior of interneurons in the spinal cord which produce a rhythmic motor discharge in the absence of descending and peripheral synaptic inputs (Ziskind-Conhaim et al., 2008) and a similar spontaneous firing occurs in tuberomammillary neurons (Taddese and Bean, 2002); fast rhythmic bursting in layer 2/3 cortical neurons is enhanced by persistent inward currents caused by Na^+ channels (Traub et al., 2003); oscillations in entorhinal cortex layer V neurons are caused by spontaneous Na^+ channel activity around the resting potential (Agrawal et al., 2001).

An intriguing aspect of voltage-gated Na^+ channels is the relationship between their inactivation characteristics and the normal resting potential in cells. Resting potential in chemoreceptor afferent neurons, for instance, is about −58 mV (Belmonte and Gallego, 1983; Donnelly, 1999; Iturriaga et al., 2007) but the half inactivation potential (i.e., the voltage where half the inactivation particles, h, are bound) for TTX-S currents is about −70 mV, meaning more than half of the Na^+ channels are not available to open and support AP generation (Rush et al., 2005). This suggests that either nature is a poor designer of channels and should have moved the inactivation potential in the positive direction or the inactivation characteristic is used to support a role different than providing the ionic basis for the rise of the AP. One possible role is due to the finite probability for the inactivation particle, h, to dissociate from its binding site, thus allowing current to flow and generating a depolarization. When averaged over a large number of channels in a relatively large structure (e.g. in a soma) this would be manifest as a net inward current whose magnitude increases with slight depolarizations, termed a persistent Na^+ current. When present in a small structure with a comparatively small number of channels (e.g. nerve terminals) the small, episodic currents contribute to membrane noise and even form the basis for AP generation in small nerve fibers (Chow and White, 1996).

2.4. Characteristics of Na^+ currents in mature chemoreceptor afferent neurons

The first intracellular recordings of chemoreceptor afferent neurons were undertaken in cat ganglia by Belmonte and Gallego using sharp electrodes (Belmonte and Gallego, 1983). Carotid bodies were harvested, intact, with the petrosal ganglia, allowing an unequivocal identification of chemoreceptor modality based on spontaneous AP activity and response to excitatory agents (Belmonte and Gallego, 1983; Belmonte et al., 1988). The high electrical resistance of sharp electrodes precluded the ability to perform voltage-clamp measurements on Na^+ channels, but some channel characteristics could be inferred from the AP waveform. All petrosal chemoreceptor neurons with a conduction velocity faster than 2 m/s demonstrated a “hump” on the falling phase of the action potential which was sensitive to the removal of extracellular Na^+ ion (Belmonte and Gallego, 1983; Gallego, 1983; Varas et al., 2003). The biphasic waveform suggests the presence of two types of Na^+