SENSORY NERVES IN THE RESPIRATORY tract are adapted to detect various elements of the physical and chemical environment of the airways and transmit this information to the central nervous system (CNS). The information that is sent from the terminals to the CNS is digitally encoded in the form of action potentials. Before an action potential is evoked, the relevant sensory stimulus must first be transduced into a depolarization of the nerve membrane. This is often referred to as the “generator potential.” If the generator potential is of sufficient magnitude, it leads to the formation of the all-or-nothing action potential via the activation of a family of voltage-gated sodium channels. The action potential is conducted along the axon until it invades the central terminal, causing neurotransmitter (usually glutamate) release into the synapse of the secondary neurons. This ultimately leads to sensations and reflex actions. If the generator potential is below the action potential threshold, it will electrically fade and no information is sent.

This review focuses on the various mechanisms by which chemical and mechanical stimuli are transduced into the generator potential. It is important to note that chemicals and mediators interacting at the afferent nerve terminal can also affect the nerve without causing a generator potential. For example, the stimulus may be “neuromodulatory” in that it changes the excitability such that it alters its response to subsequent activating stimuli. Chemicals may also interact with the terminals in a manner that leads to changes in gene expression at the cell body (i.e., neuroplasticity). These different outcomes can be activated by transduction mechanisms that are relatively independent of each other. This overview will also briefly review literature pertinent to the transduction mechanisms involved in these nonactivating stimuli.

AIRWAY SENSORY NERVE SUBTYPES

In the most general terms, the sensory nerves most often studied in the airways can be subdivided into two general categories, those that respond to the mechanical forces caused by the inflation and deflation of respiration (stretch receptors) and those that are adapted to respond to the threat of tissue damage (nociceptors). The potential threat of tissue damage may be overt trauma or more often inflammation, excessive stretch, etc. The stretch-sensitive mechanosensors can further be subdivided on the basis of their adaptation properties to a sustained inflation into slowly adapting receptors (SARs) and those that discharge action potentials on lung inflation but then rapidly adapt to the stimulus (RARs) (41, 103). The stretch receptors are classified as “A fibers” because action potentials are conducted at a fast rate along their axons (~10–50 m/s). Most nociceptors in the lung are classified as “C fibers” because they conduct action potentials at a slow velocity (~0.3–2 m/s). The nociceptive C fibers can be further subdivided into those situated deep in the lung (pulmonary C fibers) and those in the conducting airways (bronchial C fibers) (21). Regardless of their location, nociceptive C fibers in the airways are typically stimulated by capsaicin (21, 58, 91, 96, 99). In addition to these fiber types, in the guinea pig larynx, trachea, and main bronchus, a fiber has been reported that is neither a stretch receptor nor is it a capsaicin-sensitive C fiber (84). This A-type fiber appears to be rather specifically adapted to respond to mucosal touch (punctate mechanical stimulation) or...
rapid changes in pH (10). Activation of this fiber initiates cough reflexes in the guinea pig (10), although other nerve pathways are also able to evoke cough reflexes (102).

Vagal afferent nerves that do not readily fit into either the “nociceptor” or “stretch receptor” categories are those that are found to innervate the neural epithelial bodies (8, 100, 105). Anatomical studies clearly indicate that vagal A fibers innervate these structures, but neither the stimulus profile that leads to their activation, nor the consequent reflex activity has yet been worked out. Likewise, in the nasal mucosa there are solitary innervated chemoreceptor cells that defy classification until more is known about their properties (31).

The cell bodies of sensory nerves in the nose are situated in the trigeminal ganglia, whereas those in the lower airways are situated in the vagal sensory ganglia and the dorsal root ganglia (DRG). There are two vagal sensory ganglia termed the nodose and jugular (or supranodose) ganglion. Neurons in the nodose ganglion are derived embryologically from the epibranchial placodes, whereas those in the jugular ganglion (and DRG) are derived from the neural crest (3). The RAR and SAR stretch receptors, the A fibers innervating the neural epithelial bodies, and the touch-sensitive fibers in the trachea are derived from nodose neurons (10, 23, 24). Bronchopulmonary C fibers are derived from both nodose, jugular, and DRG (1, 93, 99, 100). The phenotype and activation profile of a bronchopulmonary C fiber appears to depend in large part on whether it is placodally derived (nodose neurons) or neural crest derived (jugular and DRG neurons) (15, 19, 99). The trigeminal ganglia contain a mixture of placodal and neural crest derived neurons (3), but the embryological source of the trigeminal neurons specifically innervating the nasal mucosa is as yet unknown.

**ACTIVATION: INDUCTION OF THE GENERATOR POTENTIAL**

Airway neurons respond to mechanical stimulation (punctate and stretch) as well as chemical stimulation (e.g., inflammatory mediators) by converting these analog stimuli into changes in ionic conductance across the membrane, leading to depolarizations in the membrane potential. This initial depolarization of the afferent terminal membrane is referred to as the generator potential. Unlike the action potential, the amplitude of the generator potential increases with increased stimulus intensity. Also unlike the action potential, the generator potential is not actively conducted along the axon. If this generator potential exceeds the threshold of the specific voltage-gated Na⁺ channels, it will trigger the production of “all-or-nothing” action potentials that are then conducted along the fiber to the CNS. By analogy with synaptic transmission, the generator potential is similar to the excitatory postsynaptic potential that passively invades the spike initiation segment of the nerve, where, if a threshold depolarization is met, action potential discharge is evoked. Generator potentials have been directly studied in other systems, most commonly in the crayfish stretch receptors (37) but have not been directly studied in vagal afferent nerves. It is therefore unknown whether the site at which the generator potential is initiated is spatially removed from the spike-initiation segment in the terminal of vagal sensory nerves (as the site of excitatory postsynaptic potential initiation in the dendrite is removed from the spike initiation segment in the axon hillock). In any event, the discussion below concerning activation of bronchopulmonary afferents deals mainly with the mechanisms by which various stimuli can interact directly with the afferent terminals to cause, in theory, suprathreshold generator potentials.

**Mechanical Activation**

Most airway afferent nerves are reproducibly responsive to some sort of mechanical stimulation. Punctate stimulation of the nerve terminal, by the use of von Frey fibers, activates most sensory terminals, but the sensitivity is markedly different among the different nerve subtypes. The touch-sensitive Aβ fibers in the trachea are much more sensitive to this type of mechanical stimulation than, for example, C-fiber terminals within the same tissue (84). The SARs and RARs are sensitive to the mechanical forces of tissue distension caused by lung inflation. Bronchopulmonary C fibers are relatively insensitive to the mechanical forces of eurpneic respiration but may be stimulated by mechanical forces associated with edema (85).

One can safely assume that specific accessory proteins and ion channels in the nerve terminal mediate the response to mechanical stimulation (36, 69). The identity of these airway mechanotransductive proteins is not known. The “physical hypothesis” for mechanical nerve activation is that mechanical stimulation physically alters a mechanogating protein leading to the opening of a cation channel in the terminal membrane. One particular superfamily of proteins that includes epithelial sodium channels (ENaCs), which have extensive sequence homology with the MEC family of mechanotransducing proteins in Caenorhabditis elegans (95), have attracted attention as mechanogating proteins in mammals. In mouse colonic afferent nerves, the acid-sensing ion channel-3 (ASIC3), a channel in the ENaC superfamily, has been implicated in the overall afferent response to mechanical stimulation (52). The mRNA that encodes ENaC subunits has been demonstrated in rat nodose, trigeminal, and DRG neurons (27, 28, 33), and amiloride, a nonselective inhibitor of ENaCs, caused a reduction in the mechanical activation of guinea pig lower airway afferents (11). However, mRNA for α-ENaC, a subunit critical for the formation of a viable cation current conducting protein, was not present in either DRG or nodose ganglia (27, 28). Recently, several members of the transient receptor potential potential (TRP) family of ion channels have been implicated in sensory nerve mechanotransduction (66). Two of these channels in particular, TRPA1 and TRPV4, are known to be expressed in nodose sensory nerves (106). The amount of information regarding mechanisms of sensory mechanotransduction is increasing at a rapid rate. Nevertheless, at present, the molecular basis of mechanical transduction in sensory terminals of the respiratory tract remains a proverbial black box. It would seem that the mechanism of visceral mechanotransduction is an area ripe for new discovery in the near future.

The “chemical hypothesis” of mechanotransduction is that mechanical perturbation of the tissue causes the release of substances from nonneuronal cells such as epithelial cells that subsequently activate the “mechanosensitive” nerve. Such a hypothesis could help indicate a possible function of neuroepithelial bodies, structures that have been shown in elegant immunohistochemical studies to surround populations of vagal and spinal lower airway afferents (8, 100). Mechanical deformation of the neuroepithelial bodies has been shown to evoke the release of 5-HT (79), a mediator that may activate sensory...
nerves via interactions with ionotropic 5-HT3 receptors (see Chemical Mediator Activation; Ref. 10). Studies of the mechanotransduction mechanisms in the bladder suggest that epithelial cells reproducibly release ATP in response to mechanical stimulation (54, 89). ATP then acts as the chemical transducer of mechanical stimulation by activating the nerves innervating the bladder. The RAR and SAR fibers in the guinea pig lungs are very sensitive to ATP (10). ATP evokes robust action potential discharge in these fibers via the interaction with ionotropic purinergic receptors (P2X-type) on the nerve terminals (see Chemical Mediator Activation; Ref. 10).

A defining feature of mechanoreceptors is their rate of adaptation. As mentioned above, vagal airway stretch receptors are no exception, where both rapidly and slowly adapting receptors can readily be distinguished. The transduction mechanisms underlying adaptation of vagal afferent nerves are not known. On the basis of studies in other systems, it is clear that adaptation can occur at the level of the generator potential, or at the level of spike initiation. Examples of adaptation at the level of the generator potential have long been noted in Pacinian corpuscles and crayfish stretch receptors (67, 74). A sustained indentation of Pacinian corpuscle produces a rapidly adapting generator potential and consequently a rapidly adapting action potential discharge. In this case the adaptation is due to the mechanical properties of the corpuscular end organ. When the corpuscle is stripped from the underlying nerve terminals, the sustained stimulus is met with a nonadapting generator potential (67). Alternatively, a sustained membrane depolarization, in theory, may lead to rapidly adapting spiking via intrinsic electrophysiological properties of the nerve membrane. Supporting this view are studies in which the cell bodies of rapidly adapting cutaneous mechanosensors responded to a sustained suprathreshold depolarizing intracellular current pulse with rapidly adapting action potential discharge, whereas the cell bodies of slowly adapting cutaneous mechanosensors responded to the depolarizing current step with sustained action potential discharge (39). It should be pointed out, however, that vagal mechanosensors in guinea pig trachea respond to punctate mechanical stimulation in a rapidly adapting fashion, even when, at the level of the cell body, a sustained current step often results in nonadapting action potential discharge (70).

The rapidly adapting response of RARs to lung inflation is unlikely due to intrinsic electrophysiological properties, because these nerves have been found to respond in a relatively nonadapting fashion to other stimuli such as lung deflation (41) and bronchoconstrictor agonists (5). The rapidly adapting touch-sensitive fibers in the trachea can also respond in a nonadapting fashion with different types of stimulation (70). These data would argue, therefore, that the rapidly adapting response to lung inflation is at the level of the generator potential. The RAR terminals would appear to be situated in the tissue in such a manner that sustained lung inflation leads to a generator potential that responds to the dynamic aspects of lung inflation but then rapidly fades over time during the static portion of the sustained inflation.

Chemical Mediator Activation

Many, perhaps most, chemical mediators delivered to the lungs can influence action potential discharge at afferent nerve terminals. This is often due to indirect actions of the mediators. For example, any mediator that leads to bronchospasm can indirectly lead to action potential discharge in RAR and SAR fibers. Likewise, mediators that change compliance of the lungs or cause pulmonary edema can result in activation of RAR fibers and certain C fibers (81, 85). Finally, any stimulus that causes a local inflammatory reaction can lead to vagal afferent nerve activation secondary to the release of neuroactive mediators (e.g., bradykinin, ATP, 5-HT, adenosine, acid, etc.). We will not address these indirect mechanisms of activation in this review.

Relatively few chemical mediators have been found to directly and overtly activate vagal afferent nerves in the respiratory system. Evidence for direct activation of the terminal by a chemical mediator is best obtained by recording from nerve endings within an isolated tissue environment, in which potential indirect influences can be experimentally minimized. Additional support for a direct action of a chemical mediator on the nerve ending can be obtained by using electrophysiological recordings at the sensory cell bodies retrogradely labeled from the tissue of interest. For example, in Fig. 1 we see that ATP causes activation of nodose C fibers in a segment of isolated innervated lung. In support of a direct mechanism of action, ATP also evokes a large inward current in the cell body of an isolated neuron that projects its fiber to the lungs. Results from this strategy must be cautiously interpreted because it assumes that the receptors for the chemical stimulus, in this case purinergic P2X receptors, at the terminals will also be present in the membrane of the cell body. In general we have found this assumption to be valid on the basis of our experience with capsaicin, bradykinin, 5-HT, adenosine, and ATP. Another related strategy that is useful in addressing hypotheses concerning direct- vs. indirect-acting chemical stimuli is one in which the retrogradely labeled neurons are evaluated for expression of the suspected receptor genes by using standard single-cell RT-PCR techniques.

As a rule, the nociceptive nerves are more responsive to chemical mediators than the stretch-sensitive RAR and SAR fibers or the touch-sensitive cough fibers. The C fibers are often referred to as “polymodal” fibers, because they respond to a broad range of stimuli. Nociceptive nerves throughout the visceral and somatosensory system share this polymodal characteristic.

Ionotropic receptors. Most chemical mediators that activate bronchopulmonary afferent nerves stimulate ionotropic receptors (Table 1). Ionotropic receptors are both receptors, in the traditional sense that they bind with high-affinity specific ligands, and also ion channels. When the ligand interacts with the binding site it causes a deformation in the protein, resulting in the formation of a pore through which ions pass. The ionotropic receptors in sensory nerves most often form a pore that selectively conducts cations. The cations fluxing through the pore depolarize the membrane, causing the generator potential.

Capsaicin is a vanilloid compound that stimulates nociceptive C fibers in mammalian tissues including the airways. About 10 years ago, the receptor for capsaicin was discovered to be a ligand-gated cation channel (16). This receptor is termed TRPV1 (transient receptor potential vanilloid-1). Endogenous stimulators of TRPV1 include intense heat, acidic solutions, certain endocannabinoids, and metabolites of arachi-
donic acid (16, 47, 55, 107). Although TRPV1 is accurately classified as an ionotropic receptor for certain vanilloids such as capsaicin, this ion channel can also be gated by various signaling elements initiated by the activation of metabotropic GPCRs (as discussed Metabotropic receptors). TRPV1 on vagal afferent C fibers in the lungs may also be activated by reactive oxygen species, but whether this is a direct effect or secondary to the release of other TRPV1 stimulators is unknown (86).

ATP effectively activates pulmonary C fibers in the dog and the placodal (nodose) population of C fibers in guinea pigs (46, 99). ATP is also a potent and effective stimulant of stretch receptors in the guinea pig lungs (10). In fact, although many autacoids can lead to activation of stretch receptors in the lung indirectly by causing bronchoconstriction, ATP appears to be one of the few autacoids that can directly stimulate stretch receptors. The ATP (purinergic) receptors are classified as metabotropic (P2Y receptors) and ionotropic (P2X) receptors, with a large number of receptor subtypes in each category. A pharmacological analysis reveals that the receptors responsible for initiating action potential discharge in nodose C fibers and stretch-sensitive fibers belong to the ionotropic P2X class (10). The specific subtype of P2X receptor responsible is unknown, but in both C fibers and stretch receptors it has characteristics in common with P2X2 and P2X3 receptors. Patch-clamp studies in nodose neurons support the hypothesis that some of the purinergic ionotropic receptors on visceral neurons may be heteromeric P2X2/3 receptors (20).

Serotonin has long been known to stimulate respiratory C fibers (21). Among the 13 or so subtypes of 5-HT receptors, one subtype, 5-HT3, is an ionotropic receptor. Not surprisingly, it is the 5-HT3 receptor that is responsible for 5-HT-induced action potential discharge in airway C-fiber terminals (19). Many of the classical studies on cardiopulmonary reflexes evoked by C-fiber stimulation used phenylbiguanide, a chemical now known to be a selective 5-HT3 receptor agonist, as a selective C-fiber stimulant (21).

The family of cholinergic nicotinic receptors are all ionotropic receptors. Nicotine and other nicotine receptor agonists can stimulate C fibers in certain species via interactions with nicotinic receptors (57, 61). The specific subtype of nicotinic receptors involved has not yet been worked out. Acidic solutions lead to C-fiber-mediated respiratory reflexes (62, 75). Protons can interact with ionotropic receptors, leading to the stimulation of airway afferent nerves. As mentioned above, one such receptor sensitive to acidic pH is TRPV1 (the capsaicin receptor) (16, 55). At body temperature, activation of TRPV1 requires a pH of ~5–6, raising the

![Fig. 1.](image_url)

**Table 1. Chemical stimuli that can induce action potential discharge in airway sensory nerves**

<table>
<thead>
<tr>
<th>Chemical Mediator</th>
<th>Receptor Mechanism</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Ionotropic receptors</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Capsaicin</td>
<td>TRPV1</td>
<td>55, 59</td>
</tr>
<tr>
<td>Acid</td>
<td>TRPV1</td>
<td>56</td>
</tr>
<tr>
<td>ATP</td>
<td>P2X</td>
<td>99</td>
</tr>
<tr>
<td>5-HT</td>
<td>5-HT3</td>
<td>19</td>
</tr>
<tr>
<td>Nicotine</td>
<td>nACh</td>
<td>61</td>
</tr>
<tr>
<td><strong>Metabotropic receptors</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bradykinin</td>
<td>B2</td>
<td>14, 48, 63</td>
</tr>
<tr>
<td>Adenosine</td>
<td>A1</td>
<td>42</td>
</tr>
<tr>
<td>Other</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Distilled water</td>
<td>?</td>
<td>84</td>
</tr>
<tr>
<td>Hypertonic saline</td>
<td>?</td>
<td>80</td>
</tr>
</tbody>
</table>

Many other mediators are associated have been associated with afferent nerve activation [e.g., histamine, PGE2, PGF2α (21), but the evidence is lacking for a direct mechanism of action (see text)].
question of whether this is an important mechanism in visceral organs. A decrease in extracellular pH also activates capsaicin-insensitive guinea pig tracheal A-fiber afferents and tracheal C fibers in the presence of TRPV1 antagonists, capsazepine, and indoressiniferatoxin (56), as well as lower airways C fibers in TRPV1 knockout mice (55). These data support the hypothesis that acid can activate airway afferent nerves by a TRPV1-independent mechanism(s). The details of this mechanism are unknown, although evidence suggests that certain members of the ASIC family of ion channels may be involved. This apparent ASIC-like mechanism is more sensitive to changes in pH than TRPV1. Unlike acid activation of TRPV1, activation of the ASIC-like mechanism is rapidly desensitized or “inactivated.” In biophysical studies on certain ASICs, if the pH is decreased at a slow rate (94), inactivation mechanisms are set in even before the activation of the channel occurs. Likewise in vagal afferent A-fibers in the trachea, a relatively rapid decrease in pH is required to discharge action potentials via this TRPV1-independent mechanism (56).

Metabotropic receptors or GPCRs. Agonists that interact with G-protein-coupled receptors (GPCRs) can also lead to generator potentials that evoke action potential discharge in airway sensory nerves. Again, this mechanism for activation appears to be limited primarily to polymodal nociceptors. Examples of two GPCRs capable of causing action potential discharge in bronchopulmonary C fibers are bradykinin B2 receptors (63) and adenosine A1 receptors (42).

The mechanism by which stimulation of a GPCR is transduced to membrane depolarization is more complex than the simple ionotropic mechanisms, and the details of this signaling have not been completely worked out. At some point the signaling through the G-protein must lead to the opening or closing of specific ion channels that lead to a net membrane depolarization. One ion channel that appears to be involved in the coupling of Gq-linked receptors (such as the bradykinin B2 receptor) is the capsaicin receptor TRPV1 (14, 92).

There are at least two signaling pathways initiated by Gq-coupled receptors that may lead to the opening of TRPV1. Under resting conditions TRPV1 is inhibited by phosphatidylinositol-4,5-bisphosphate (17). Activation of phospholipase C via the Gq-coupled mechanism leads to the enzymatic cleavage of phosphatidylinositol-4,5-bisphosphate and a loss of the TRPV1 inhibition. The second pathway through which Gq-coupled receptors can activate TRPV1 is via the production of lipoxygenase products of arachidonic acid. In this scenario the increase in intracellular calcium leads to the activation of phospholipase A2 and the liberation of intracellular arachidonic acid. The lipoxygenation of arachidonic acid produces messengers that can then stimulate TRPV1 (92). In airway C fibers, pharmacological blockade of TRPV1 inhibits bradykinin B2-induced action potential discharge (14). Inhibition of lipoxygenase enzymes also inhibits the bradykinin response in these cells (14).

Bradykinin can evoke action potential discharge in airway C fibers, even when TRPV1 is pharmacologically blocked (14, 63), or in mice in which TRPV1 has been genetically knocked out (55). The non-TRPV1 component of Gq-coupled activation of C fibers may involve the gating of certain calcium-activated chloride channels (63). Primary afferent neurons, unlike CNS neurons, contain relatively large concentrations of chloride. This is due to the expression and activity of a particular (Na+)-(K+)-(2Cl−) cotransporter, namely NKCC1 (25, 38). This protein pumps two chloride anions into the cell in exchange for a sodium and potassium cation. The resulting effect is that the equilibrium potential for chloride is positive relative to the resting potential. Thus, if chloride channels are opened in primary afferent nerves, the chloride ions flow out of the terminal, causing membrane depolarization. In CNS neurons (which do not express NKCC1), the equilibrium potential is negative relative to resting potential. A substantial component of the net depolarization caused by bradykinin B2 receptor activation in nodose neurons is via chloride efflux (78). In airway C-fiber terminals, the TRPV1-independent component of bradykinin-induced action potential discharge is blocked by inhibitors of calcium-activated chloride channels (63). Odorants act via GPCRs to stimulate olfactory neurons (9). Like the vagal sensory neurons, it appears that a major component of the GPCR transduction mechanism by which odorants activate olfactory neurons is also via chloride channel activation (82). These data indicate that NKCC1 may be another potential therapeutic target for drugs aimed at decreasing activity of airway afferent nerves.

Recently, in DRG neurons, bradykinin has been found to increase cellular calcium through interaction with both TRPV1 and another TRP channel, namely TRPA1 (4). The role of TRPA1 (an ionotropic receptor for the active ingredients in mustard and garlic) in the activation of bronchopulmonary afferent nerves is not known.

Osmolarity. Water is a powerful activator of airway afferent nerves (32). Hyperosmolar solutions also stimulate airway afferent nerves (80). Despite the breadth of pharmacological and molecular information linking airway afferent chemical activation to chemical mediators, the mechanisms underlying afferent activation by hypertonic and hypertonc solutions are poorly understood. It is uncertain whether changes in osmolarity activate neurons because of the presence of a specific osmosensor or whether activation is due to mechanical forces of tissue swelling or shrinkage caused by the movement of water or to subsequent changes in electrochemical gradients. Simply reducing the extracellular chloride concentration, for example with a solution of isotonic dextrose, stimulates airway afferent nerves (32, 88), probably because of the chloride gradient due to the activity of NKCC1 as discussed above. Indeed, the afferent nerve activation caused by a low-chloride solution can be inhibited by the chloride pump inhibitor furosemide (32, 88).

TRPV4 and a variant of TRPV1 have been identified as possible contributors to osmosensitivity (2, 73). These TRP channels nonselectively permit the flow of cations and, as such, are capable of action potential initiation once activated. In the guinea pig, jugular capsacin-sensitive C fibers and especially the capsacin-sensitive Aδ fibers are the fiber types most sensitive to hypertonic solutions (80). Whether TRP channels are involved in these responses has not yet been investigated.

Temperature

The effect of temperature on airway afferents has not been as extensively studied as have cutaneous temperature sensors. Studies have shown that cutaneous C fibers are activated by absolute temperatures, not by the rate of temperature change, and that stimulus history can have either a sensitizing or
NEUROMODULATION

In the context of the present review, neuromodulators include those stimuli that act directly on the nerve terminal to increase or decrease the excitability of the afferent nerves but do not overtly activate the nerve.

Increased Excitability

With the few exceptions discussed above (bradykinin, adenosine), most mediators and autacoids fail to directly activate lower airway afferent nerves (although this may not be the case for nasal afferents). Many of these mediators bind to specific GPCRs at the nerve terminal, leading, instead, to a nonspecific increase in the electrical excitability of the nerve (Fig. 2). This can occur secondarily to phosphorylation of certain voltage-gated sodium channels, leading to an increase in their conductance characteristics. Autacoids can also cause a generalized increase in membrane resistance via the inhibition of certain potassium ion channels. Alternatively, neuromodulators may lead to selective increases in the excitability of specific activator pathways (e.g., phosphorylation of TRPV1).

The subcellular mechanisms by which neuromodulation occurs are diverse and complex and are beyond the scope of this brief review. One example of this complexity is illustrated by PGE2 (Fig. 3), which sensitizes primary afferent neurons by at least four known mechanisms following Gs activation. In general Gs is linked to adenyl cyclase so that its activation leads to an increase in cAMP. Activation of adenyl cyclase in vagal neurons, by either PGE2 or forskolin, leads to inhibition of the calcium-activated K+ current known as the AHPslow current (101), thus decreasing the duration of the afterhyperpolarization (AHP) and increasing the electrical excitability of the nerve. In addition, PGE2-induced cAMP increases the hyperpolarization-activated cation-conducting If current in nose and trigeminal neurons in a protein kinase A-independent mechanism (49), also limiting the duration of the AHP. Increases in cAMP can also increase TRPV1 activity by decreasing the rate of TRPV1 desensitization (72). Finally, PGE2-induced cAMP activates PKA, which phosphorylates tetrodotoxin-resistant sodium channels in visceral afferent nerves (35) including lung-specific vagal neurons (58), increasing voltage sensitivity and conductance of these channels.

In many cases, neuromodulators cause a small amount of membrane depolarization (~1–4 mV) accompanied by an increase in membrane resistance. Neuromodulators such as PGE2 and histamine inhibit resting potassium “leak” currents, causing an increase in membrane resistance and a gradual buildup of cations within the cell, thus producing the minor depolarization. Perhaps more importantly, this will also lead to an increase in membrane resistance, theoretically resulting in an increase in the amplitude of generator potentials produced by a given amount of activating stimulus (e.g., mechanical perturbation, capsaicin, acid). An increase in the rate and amplitude of the generator potential will, in turn, cause a change in stimulus threshold and/or an increase in the peak frequency of action potential discharge. PGE2 inhibition of IK currents appears to be cAMP and PKA dependent in rat DRG neurons (30). In ferret vagal afferent neurons IK currents are inhibited by histamine in an H1-sensitive mechanism (50). The precise identity of the K+ channels that contribute to the resting current in airway afferent nerve terminals is unresolved.
but several members of the voltage-gated potassium channels (Kv channels) have been implicated in regulating excitability of visceral sensory neurons (34, 90).

**Decreased Excitability**

Theoretically, any stimulus that decreases membrane resistance of the afferent nerve, especially if this is accompanied by a membrane hyperpolarization, can lead to a generalized decrease in excitability of that nerve. Agents that open K⁺ channels are therefore potentially downregulators of airway afferent function. Indeed, pharmacological openers of Ca²⁺ channels are therefore potential downregulators of airway afferent function. Agents that open K⁺ channels (Maxi-K type) have been found to decrease airway afferent nerve excitability (104), and openers of A-type potassium channels have been associated with decreased excitability of other visceral afferent nerves (68). Mediators that lead to a decrease in the activity of voltage-gated sodium channels would not inhibit generator potential amplitude but would decrease the ability of the generator potential to generate action potentials. Other than local anesthetics and certain toxins, there has been little published on the selective downregulation of sodium channels in airway sensory nerves.

Nociceptin, the endogenous agonist of the metabotropic "opioid-like" receptor NOP1, inhibits airway afferent nerve activity in a somewhat selective manner. This peptide has been shown to inhibit cough elicited by capsaicin, acid, and mechanical stimulation (6, 64, 71). At a mechanistic level, nociceptin was demonstrated to selectively inhibit capsaicin-induced calcium increases in isolated neurons (51) and to inhibit the TRPV1 component, but not the non-TRPV1 component, of the acid-induced inward current in airway jugular C-fiber neurons (64). The precise mechanism by which nociceptin reduces TRPV1 functions in airway afferents is unknown, although it is inhibited by a selective NOP1 antagonist (51).

**TRANSCRIPTIONAL REGULATION**

Airway afferents in the mature adult animal are not static with respect to gene expression and protein production. In particular external influences such as disease, injury, and inflammation are able to cause changes in expression of various genes involved in the production of neuropeptides, neurotransmitters, and various ion channels. These long-lasting changes in nerve function are often referred to as neuroplasticity. The plastic nature of the afferent nervous system presents a major challenge for those interested in understanding how sensory nerves contribute to inflammatory airway diseases, as it is apparent that the nerves need to be studied within the context of the inflammatory state.

Primary afferent nerves are usually erroneously depicted with relatively short peripheral and central processes. It is important to keep in mind that any mechanism in the airways that leads to changes in gene transcription must incorporate the fact that the cell nucleus is actually a long way from the terminals in the airways. For example, a neuron with a cell diameter of 30 μm situated in the nodose ganglion has its terminal >10,000 cell diameters away in the wall of the human bronchus; to put it another way, if the sensory cell body were imagined to have the dimensions of a tennis ball, the nerve terminal in the airway would be some 600–700 m away. Two mechanisms that can signal across such long distances involve a family of trophic factors called neurotrophins and conducted action potentials.

**Neurotrophins**

Neurotrophins are polypeptides that support growth, differentiation, and survival of neurons in developing and adult nervous systems. The prototypical neurotrophin is nerve growth factor (NGF), but this family also includes brain-derived neurotrophic factor, neurotrophin 3, neurotrophin 4, and neurotrophin 4/5. Neurotrophins act on a family of receptor tyrosine kinases (Trk); NGF preferentially activates Trk-A receptors, brain-derived neurotrophic factor and neurotrophin-3 are selective for Trk-B receptors, and neurotrophin-4 is selective for Trk-C receptors (reviewed in Ref. 13) (65). Recently there has been interest in the potential role of neurotrophins in airway pathophysiology (77). The sources of neurotrophins in the airway include infiltrating cells, fibroblasts, smooth muscle, and epithelium (83). NGF is elevated in the
serum of allergic individuals (7). Respiratory viral infections, in particular, are associated with elevation of NGF in the airways (98).

The binding of neurotrophins to the high-affinity Trk receptors causes autophosphorylation of receptor tyrosine residues, followed by uptake of the ligand-receptor complex and retrograde axonal transport to the cell body (22), and possibly beyond into the CNS (40). In the cell body the ligand-receptor complex activates various “effector” molecules such as extracellular signaling regulated kinases and Cre element binding protein (see review in Ref. 43), which induce multiple genomic effects. One effect of neurotrophic factors in airway afferent plasticity is their effect on neuropeptide content. For example, overexpression of NGF in mouse airway epithelium leads to a tremendous increase in substance P content of airway nerves (44). Allergic (18) and viral (12) inflammation causes the production of neuropeptides in large-diameter (A-fiber) guinea pig vagal airway afferent neurons. These neurons, which normally do not produce neuropeptides, include those that supply the trachea with touch-sensitive cough fibers and intrapulmonary stretch receptors. Consistent with a neurotrophic-like mechanism for this effect was the observation that when the vagal axons attaching the terminals to the cell bodies were severed, the allergen-induced induction of neuropeptide production in A-type neurons was inhibited. Moreover, this type of neuroplasticity caused by allergic and viral inflammation is mimicked by local injection of NGF into the guinea pig (45) and mouse airways (26).

**Use-Dependent Mechanisms**

Action potentials arising at the nerve terminals invade the cell somal membrane on their way to the central terminals. The somal membrane contains voltage-gated calcium channels, such that the action potential will lead to the influx of calcium through voltage-gated calcium channels. This provides a mechanism by which information at the terminal may be transmitted to the nucleus. It is not known to what extent the actual activity of an airway afferent nerve impacts on its own gene regulation. Use-dependent genomic regulation has been demonstrated in mouse DRG neurons (53) but has yet to be shown in airway-specific neurons.

**CONCLUSIONS**

The general model for action potential initiation in airway sensory nerves involves a stimulus causing a generator potential via the opening of cation channels in the plasma membrane. Chemical activators evoke generator potentials by interacting with either ionotropic receptors or through the action of GPCRs. At least some of the activating GPCRs signal the opening of ion channels (e.g., TRP channels and certain calcium-gated chloride channels) through the action of phospholipase C and increases in intracellular calcium. Mechanical activation of afferent nerves, by inference from more primitive organisms, likely employ specialized mechanically gated ion channels; however, nearly nothing is yet known about the nature of these channels in respiratory afferent nerves. Release of chemical mediators from mechanically sensitive nonneuronal cells may also contribute to the net effect of mechanical perturbations on action potential discharge in some airway mechanosensors.

Neuromodulators can alter the electrical excitability of airway afferent nerves in the absence of overt activation. The neuromodulators usually act via GPCRs and affect ion channel activity via several signaling mechanisms. In addition, other, independent, mechanisms of airway afferent activation can result in changes in gene expression involved in neuroplasticity at the level of the cell nucleus. Although this review touched with broad strokes on some of the highlights of these transductions mechanisms, it is clear that much remains to be learned before we understand how the airway environment is recognized and transduced into meaningful information by the sensory nervous system.

**REFERENCES**


18. Chuaychoo B, Hunter DD, Myers AC, Kollari K, and Undem BJ. Allergen-induced substance P synthesis in large-diameter sensory neu-}


21. Cappiello L, Bautista DM, and Jordt SE. The capsaicin receptor is a}


24. Carr MJ, Kollari K, Meeker SN, and Undem BJ. A role for TRPVI in bradykinin-induced excitation of vagal airway afferent nerve termi-}


TRANSDUCTION MECHANISMS IN AIRWAY SENSORY NERVES


