Pharmacology of the Equine Tracheobronchial Tree

N. Edward Robinson, B. Vet. Med., PhD, MRCVS

The pharmacological management of tracheobronchial disease primarily involves the use of bronchodilators and anti-inflammatory agents. The most useful bronchodilators are quaternary ammonium anticholinergics, such as ipratropium, and β₂-adrenergic agonists. Bronchodilators are most effective when delivered as aerosols by one of the newer devices designed for use in the horse. Anti-inflammatory therapy involves the use of corticosteroids. Inhaled steroids are gaining popularity because they may cause few side effects. Other anti-inflammatory compounds such as inhibitors of lipoxygenase synthesis have yet to be proven useful in the horse. Blockers of specific mediator receptors, e.g., antihistamines, are rarely useful because airway obstruction involves a cascade of mediators. Authors’ address: Dept. of Large Animal Clinical Sciences, G-321 Veterinary Medical Center, Michigan State University, East Lansing, MI 48824-1314. © 1997 AAEP.

1. Introduction

The treatment of diseases of the tracheobronchial tree involves the use of drugs that, in many cases, are either agonists or antagonists of physiological receptors. This paper briefly reviews receptor physiology, describes the receptor families that are important in the airways, and discusses the therapeutic usefulness of drugs that either modify receptor function or alter the production of mediators that bind to these receptors.

2. Receptors

Hormones, autacoids (inflammatory mediators), and neurotransmitters are endogenous ligands for the receptors that regulate the function of the tracheobronchial tree. Activation of receptors initiates a sequence of responses that can result in changes in bronchomotor tone, mucus secretion, bronchial blood flow, inflammatory cell recruitment, or fluid and electrolyte transport across the airway epithelium.

Although there is a vast array of endogenous ligands of differing chemical structure binding to many types of receptors, and although there are many types of physiological effects evoked by these ligands, receptors share a limited number of intracellular pathways to evoke a response within the cell. The use of a limited number of transduction pathways allows for integration of responses when several receptors are activated simultaneously. In addition, there are interactions between transduction pathways so that activation of one pathway can inhibit or activate another pathway. Receptors can be assigned to one of several functional classes. Some, such as the receptors for peptides that regulate growth and development, are membrane-bound protein kinases that act by phosphorylating other proteins. Receptors for some neurotransmitters (e.g., the nicotinic receptor for acetylcholine) are ligand-gated ion channels. Activation of this type of receptor rapidly alters the ionic...
environment within the cell. Receptors for steroid hormones are soluble DNA-binding proteins that regulate gene expression.

From a therapeutic viewpoint in the horse, the most important receptors involved in regulation of the tracheobronchial tree are those that are coupled to guanosine triphosphate (GTP)-binding proteins. These G proteins are present on the inner face of the plasma membrane. Activation of a receptor facilitates the binding of GTP to a G protein, and this in turn regulates the activity of enzymes (e.g., adenylyl cyclase), ion channels, or transport proteins. On a single cell, receptors for a variety of ligands may all interact with the same G protein. This allows for an integrated cellular response when several inflammatory mediators or neurotransmitters are acting on the cell simultaneously. Alternatively, a single ligand may bind to several subtypes of receptors that are each coupled to a different G protein so that a variety of responses is initiated simultaneously (Fig. 1).

3. Second Messengers
The effect of many receptors on cellular function involves the generation of a limited number of second messengers. Several receptors and their associated G proteins may all affect the concentration of a single second messenger within the cell, thus providing a further method of integration of responses when a cell is exposed simultaneously to several mediators, etc.1

There are two very important second messenger pathways, cyclic adenosine monophosphate (cAMP) and Ca2+ (Fig. 2). The intracellular level of cAMP is controlled by the activity of adenylyl cyclase, which is regulated by receptors coupled to Gs or Gi proteins. Stimulatory G proteins (Gs) activate and inhibitory G proteins (Gi) inhibit adenylyl cyclase, which catalyzes the formation of cAMP from adenosine triphosphate (ATP). In addition, intracellular levels of cAMP also depend on the activity of phosphodiesterase enzymes that catalyze the breakdown of cAMP to 5′-AMP. cAMP exerts its regulatory effects by activating AMP-dependent protein kinases that catalyze the phosphorylation of intracellular proteins. β-adrenergic agonists, e.g., clenbuterol, exert their effect by means of an increase in cAMP.

The level of intracellular Ca2+ is regulated by the opening of Ca2+-specific channels in the cell membrane and by Ca2+ release from intracellular stores. The opening of channels is controlled by electrical

---

**Fig. 1.** G-protein-coupled mechanisms whereby mediators, hormones, or neurotransmitters (ligands) can interact to affect cellular function: (1) ligands may each bind to their own receptors (A and B), which are coupled to the same G protein that can initiate a change in the level of a second messenger; (2) ligands may bind to receptors (C and D) that are each coupled to different G proteins (Gc and Gd) that have opposing effects on an enzyme that regulates levels of a second messenger; (3) two second messengers may have opposing effects on a physiological function; (4) a single ligand (E) may bind to two (E1 and E2) or more receptor subtypes, each of which is coupled by its own G protein to a cellular function; (5) Second messengers can regulate the coupling of receptors to cellular function at several levels.
depolarization, G proteins, cAMP-dependent protein kinases, and the intracellular concentration of ions such as K\(^+\) or Ca\(^{2+}\) itself. Release of Ca\(^{2+}\) from intracellular stores is initiated by inositol 1,4,5-triphosphate (IP\(_3\)), which is formed from the membrane phospholipid phosphatidylinositol 4,5-biphosphate when the enzyme phospholipase C is stimulated by a receptor-activated G protein. Acetylcholine causes bronchospasm by means of the latter pathway.

4. Regulation of Receptors
Continuous activation of a particular receptor can lead to a decreased response to agonist administration. This phenomenon is known as downregulation or desensitization, and one of the best examples is the loss of the bronchodilator response to \(\beta\)-adrenergic agonists that follows their excessive use. When the desensitization is limited to the receptor that has been activated, the phenomenon is known as homologous desensitization. It can be caused by receptor phosphorylation, internalization, or destruction. Heterologous desensitization occurs when activation of one receptor leads to downregulation of several types of receptors. Usually these receptors exert their effects by means of a common pathway, and it is in this pathway that downregulation occurs. Generally, removal of the ligand that is causing desensitization results in quite rapid restoration of sensitivity.

5. Muscarinic Receptors
Muscarinic receptors are responsible for the autonomic effects of acetylcholine (ACh), which is released from the postganglionic fibers of the parasympathetic nervous system. ACh is formed in the nerve terminals, released in response to an action potential, and then rapidly inactivated by acetylcholinesterase. In the airways, parasympathetic nerve fibers supply the airway smooth muscle from the trachea to the alveolar ducts and also ramify in the lamina propria, especially around submucosal glands.

A. Smooth Muscle
Airway smooth muscle is richly supplied with muscarinic receptors. When the parasympathetic nervous system is activated, ACh is released and binds to M\(_3\)-muscarinic receptors. These receptors are coupled to the G\(_{q/11}\) protein, activation of which leads to formation of IP\(_3\) and release of Ca\(^{2+}\) from intracellular stores. This increase in the intracellular concentration of Ca\(^{2+}\) leads to smooth muscle contraction. Contraction of airway smooth muscle in response to the release of ACh and activation of M\(_3\) receptors is the primary mechanism of broncho-
spasm in the horse. In the healthy horse, tonic parasympathetic nerve activity is insufficient to cause a measurable level of airway smooth muscle contraction, but in the presence of airway inflammation, muscarically mediated bronchospasm is an important cause of airway obstruction.\(^6\)\(^-\)\(^8\)

Even though it is well accepted that activation of M2-muscarinic receptors is responsible for smooth muscle contraction, airway smooth muscle of many species also expresses M2 receptors. In some species, there are more M2 than M3-muscarinic receptors.\(^9\) The M2-muscarinic receptor is coupled by means of G\(_i\) so that its activation leads to a decrease in the intracellular concentration of cAMP (Fig. 2). When both M3 and M2 receptors are activated, acetylcholine initiates contraction by releasing Ca\(^{2+}\) and inhibits relaxation by preventing an increase in cAMP.\(^10\)\(^,\)\(^11\)

**B. Parasympathetic Nerves**

Muscarinic receptors also occur on nerves, and one receptor subtype is of particular interest with regard to inflammatory airway diseases. Postganglionic fibers of parasympathetic nerves possess M receptors, the function of which is to provide negative feedback that regulates the release of ACh. When ACh is released, some binds to and activates these prejunctional M receptors. This leads, by means of mechanisms that remain to be clarified, to a decrease in the release of ACh. The efficacy of this inhibition is demonstrated by the fact that the blockade of this receptor in vitro increases the release of ACh approximately threefold to fivefold.\(^12\) In most species, this prejunctional receptor is of the M2 subtype, but this remains to be clarified in the horse.\(^13\)

The prejunctional M2 receptor has become of clinical interest because its function is diminished in many experimental models of airway inflammation.\(^14\) For example, polycationic substances, such as major basic protein, which is released from eosinophils during hypersensitivity responses, are natural antagonists of the M2 receptor.\(^15\)\(^,\)\(^16\) Further, the influenza virus, through its neuraminidase activity, blocks M2-receptor function.\(^17\) This loss of M2 function may be a mechanism that enhances the release of ACh and causes airway hyperresponsiveness in several types of inflammatory airway disease. However, attempts to find dysfunction of the prejunctional muscarinic receptor in horses with heaves have not met with success.\(^18\)

**C. Mucus Apparatus**

Airway epithelial cells and submucosal glands express M1, M2, and M3 receptors. It is likely that these receptors are activated by ACh of neural origin because parasympathetic nerves occur in association with the surface epithelium and submucosal glands. Activation of the vagus nerve leads to an increase in the rate of mucus secretion from both goblet cells and glands and to an increase in the rate of water transport toward the airway lumen.\(^19\) The administration of cholinergic agonists leads to an increase in mucus secretion\(^20\) and in the rate of beating of cilia.\(^21\)

Activation of the parasympathetic system is responsible for increases in the rate of mucus secretion and its transport up the tracheobronchial tree following inhalation of irritants. However, tonic parasympathetic activity does not seem to be necessary for basal mucociliary function. In the species studied, the administration of most anticholinergic agents does not alter basal mucociliary activity. However, atropine is an exception to this rule. Atropine depresses mucociliary function, but the mechanism whereby this occurs is unknown. \(^1\) Ipratropium bromide, a similar nonspecific anticholinergic agent, does not affect mucociliary function. Because it can reduce airway secretions, atropine is a very useful premedication before anesthesia.

**D. Muscarinic Agonists**

Although muscarinic agonists are not used therapeutically for respiratory disease, they may be ingested accidentally. They have adverse effects on respiration by causing bronchospasm and increased mucus secretion. Cholinergic agonists include the analogs of ACh, such as methacholine and carbachol. The cholinesterase inhibitors such as neostigmine or the organophosphates, by preventing the breakdown of ACh, also act as muscarinic agonists.

**E. Muscarinic Antagonists**

The muscarinic antagonists currently used for treatment of airway disease in horses are nonselective, that is, they block all receptor subtypes. In the respiratory system, the primary use of muscarinic antagonists is for the relief of bronchospasm. This occurs when the M3 receptor on the airway smooth muscle is blocked. Muscarinic antagonists are effective as bronchodilators only when bronchospasm is a result of activation of the M3 receptor by ACh. In horses with inflammatory airway disease, this is usually the case, so this class of drugs is quite effective for bronchodilation.\(^8\)\(^,\)\(^22\)

The parasympathetic system plays a major role in the regulation of function in many organs. For this reason, the systemic administration of muscarinic antagonists has undesirable side effects, especially reduction of gastrointestinal motility. For relief of bronchospasm, muscarinic antagonists are best administered by nebulization directly into the airways. Atropine is absorbed from the airways into the blood, and systemic effects can arise following aerosol administration.

The quaternary ammonium muscarinic antagonists have the advantage that they are very poorly absorbed from the respiratory tract and also do not cause drying of respiratory secretions.\(^23\) This class of compounds includes ipratropium bromide (Atrovent\(^\text{®}\)), oxitropium bromide, and tiotropium bromide. Ipratropium bromide is an excellent bronchodilator in horses with chronic obstructive pulmonary disease (COPD).\(^7\) Inhalation of 1–3 µg/kg of ipratro-
6. Adrenergic Receptors

Adrenergic receptors are activated by norepinephrine (NE), which is released from the postganglionic fibers of sympathetic nerves, and by epinephrine (E), which is released from the adrenal medulla and reaches the lungs via the circulation. Sympathetic nerve fibers occur throughout the airways but are primarily associated with the plexus of bronchial blood vessels that underlies the epithelium. The topography of sympathetic nerves in horse airways matches the topography of sympathetic nerves in horse airways would be a cranial trachea, in which sympathetic activation causes relaxation. The major function of the sympathetic nervous system in horse airways would be to maintain bronchial function. In horses, sympathetic activation results in relaxation. This leads to upper airway obstruction, which can mask any beneficial effect on the bronchi. For this reason, we have examined the effect of aerosol administration of xylazine, a bronchodilator effect of xylazine on bronchi. No such compound is currently available for therapeutic use.

B. β Receptors

In human medicine, β-adrenoceptor agonists are the most widely used agents for treatment of obstruction of the tracheobronchial tree. The β-adrenoceptors are coupled by means of Gs, to adenylyl cyclase, and their activation results in an increase in the intracellular concentration of cAMP. Recent evidence indicates that, in addition to their effects on cAMP, β-adrenoceptors may also be coupled directly to K+ channels. Activation of β-adrenoceptors in the heart results in an increase in heart rate. This effect is mediated by the β2-adrenoceptor, which has a high affinity for NE but a low affinity for E. The beneficial effects of catecholamines on airway function are mediated by the activation of β2-adrenoceptors, which have a high affinity for E and a lesser affinity for NE. In the airways, β2-adrenoceptors are widespread, being present on the epithelium and submucosal glands and the smooth muscle. Activation of these receptors relaxes airway smooth muscle, stimulates mucus secretion and ciliary function, and inhibits release of mediators from mast cells. Isoproterenol is the prototype selective β-adrenoceptor agonist. However, its accelerator ef-
factors on the heart are so potent in horses that it is not clinically useful as a bronchodilator.

1. $\beta_2$-Adrenergic Agonists

Selective $\beta_2$ agonists retain the useful effects of isoproterenol in the airways but have much less effect on cardiac function. A wide variety of these agonists is available for use in human medicine, and they can all be used for relief of bronchospasm in horses. Ideally $\beta_2$ agonists are administered by aerosol in order to avoid any of the side effects such as sweating, nervousness, and increased heart rate. Because of the lack of convenient devices for aerosol administration to horses, clenbuterol (Ventiluplin) was developed as a $\beta_2$ agonist for systemic administration. Outside the U.S., the recommended dose of clenbuterol is 0.8 µg/kg q 12 h. Trials in the U.S. revealed that this dose is effective in only 25% of horses with COPD and that up to 3.2 µg/kg may be required in some horses. The dose must be increased gradually [by 0.8(µg/kg)/week] in order to avoid the undesirable side effects, and the maximal dose of 3.2 µg/kg should not be exceeded.

Recently the development of devices for the administration of drugs directly into the airways of horses has stimulated an interest in the aerosol delivery of $\beta_2$ agonists. Any of the drugs available for use in humans can be used in horses, but it is important to remember that these drugs may be more potent in horses than in humans. Simply multiplying the recommended human dose by five or ten may result in a dose that will cause side effects in a horse. To date, only two $\beta_2$ agonists have been extensively investigated for use in the horse—pirbuterol (Maxair®) and albuterol (Ventolin®). In horses with COPD, 600 µg of pirbuterol (three actuations of the metered-dose canister) results in bronchodilation. Relief of bronchospasm is immediate and persists for 1–2 h. Albuterol has a similar speed of onset and duration of action when given at dose of 360 µg (three canister activations).

When any drug is used as an aerosol, the efficacy of the compound will vary from horse to horse. This is due to (1) the cause of bronchospasm, (2) variations in deposition caused by the anatomy of the upper airway and tracheobronchial tree, (3) the tolerance of the horse for the delivery device, (4) the familiarity of the personnel with the delivery device, and (5) the response of the individual to the drug. With regard to the latter, many mediators released in inflammation can cause bronchospasm either by direct effects on smooth muscle or by synergizing with cholinergic responses of airway smooth muscle. The $\beta_2$ agonists, by increasing cAMP, cause a functional antagonism of bronchospasm that is effective in opposing the effects of all other mediators and neurotransmitters. $\beta_2$ agonists are least effective in opposing bronchospasm induced by release of ACh, which is a major cause of bronchospasm in the horse with COPD. This is because ACh activates smooth muscle $M_2$-muscarinic receptors, which are coupled by means of $G_1$ to inhibit the increase in cAMP initiated by the $\beta_2$ agonist.

$\beta_2$ agonists should be given to effect. Begin by using the recommended dose for horses. If the dose for horses is unknown, use three times the human dose for several administrations separated by 1–2 h and then gradually increase the dose. Undesirable side effects, such as nervousness and sweating, will limit the dose that can be used. Because of the importance of cholinergically mediated bronchospasm in horses, a combination of a $\beta_2$ and an M3 antagonist, e.g., ipratropium, should provide the greatest relief of bronchospasm.

The duration of action of $\beta_2$ agonists is variable. It is recommended that clenbuterol be given twice daily. In humans, pirbuterol and terbutaline (Brethine®) are short acting (1–2 h), albuterol and fenoterol (Berotec®) have a longer duration (up to 4 h), and the newer drug salmeterol (Serevent®) has a 12-h duration of action. In horses, pirbuterol and albuterol have the same duration of action (1–2 h).

Ephedrine is included in remedies for COPD. It is an agonist of both $\alpha$ and $\beta$ receptors and also promotes the release of NE from sympathetic neurons. It has bronchodilator activity but is a potent central nervous system stimulant.

2. Concerns About $\beta_2$-Adrenoceptor Agonists

In human medicine, there has been a concern about deaths associated with the frequent use of $\beta_2$ agonists. The evidence to support this concern is under debate. Because of the inherent difficulties in medicating horses by inhalation, it is unlikely that overuse will ever become a problem, and the concerns raised about treating humans should not deter the use of these drugs in horses.

We have recently demonstrated that, in vitro, $\beta_2$ agonists consistently facilitate the release of ACh from airway parasympathetic nerves. Theoretically, this action of $\beta_2$ agonists should oppose their bronchodilator effect on airway smooth muscle. In the whole animal, however, the bronchodilator effect of $\beta$ agonists on smooth muscle greatly outweighs any effects arising from the tendency to augment ACh release.

C. $\beta$-Adrenergic Blockers

Adrenergic blocking agents have no part in the treatment of airway disease. If they are used for other conditions, one should be aware that they can have deleterious effects in the airways of horses with chronic airway disease. When the $\beta$-blocker propranolol is administered to COPD-affected horses, it increases the severity of airway obstruction, probably by blocking $\beta_2$ adrenoceptors on smooth muscle.

7. Phosphodiesterase Inhibitors

Methylxanthines, especially theophylline and its derivative aminophylline, have been used as bronchodilators in horses. The bronchodilator properties of these drugs have traditionally been associated with
increase in intracellular Ca$^{2+}$ concentration or on the coupling of Ca$^{2+}$ to smooth muscle contractile elements, are important.

Although aminophylline is an effective bronchodilator, it has a narrow therapeutic margin and its effects vary greatly from horse to horse. The side effects of aminophylline are largely due to CNS excitement and include nervousness and tachycardia. Aminophylline is administered to horses either orally or intravenously at a dose of 5–10 mg/kg q 12 h.

Theophylline and aminophylline are nonspecific inhibitors of cAMP and cGMP phosphodiesterases. Recently, specific inhibitors for phosphodiesterase isoenzymes have been developed. In the treatment of airway disease, inhibitors of isoenzyme families III and IV are likely to be most useful because they have less effect in the central nervous system. Inhibitors of phosphodiesterase III, such as milrinone, relax airway smooth muscle. Inhibitors of phosphodiesterase IV, e.g., rolipram, relax smooth muscle and also inhibit release of inflammatory mediators.

The drugs discussed up to this point exert many of their effects on airway smooth muscle. In equine airway disease, bronchospasm is usually a result of airway inflammation, and therefore modulation of the inflammatory response is an important therapy. Drugs that block the effect of specific mediators or prevent the production of mediators therefore are also effective in treating airway disease. It is important to realize that there is redundancy in the effects of mediators of the inflammatory response. That is, several mediators have the same effects, e.g., histamine, leukotriene D$_4$, and serotonin can all cause bronchospasm. For this reason, blockers of specific receptors, e.g., the classical antihistamine drugs, are less likely to be effective than drugs such as corticosteroids that block the mediator cascade.

8. Histamine

Histamine is synthesized in many tissues, but its primary source in the respiratory system is mast cells where it is synthesized and stored. Histamine release from mast cells occurs under several circumstances. In allergic reactions, histamine is released when antigen bridges IgE molecules that are bound to the mast cell surface where they function as receptors. Histamine also can be released when the airways are exposed to cold air and to other physical stimuli. Histamine exerts its effects by binding to H$_1$, H$_2$, or H$_3$ receptors. Histamine H$_2$ receptors are coupled by means of G protein and IP$_3$ to an increase in intracellular Ca$^{2+}$. Activation of these receptors causes bronchospasm, an increase in vascular permeability, and hyperesthesia of sensory nerves. In addition to its direct effect on airway smooth muscle, histamine also contributes to bronchospasm by facilitating the response to activation of parasympathetic nerves.

Classical antihistamines are antagonists of the H$_1$ receptor. First-generation antihistamines include ethanolamines, e.g., diphenhydramine; ethylendi- amines, e.g., tripelennamine and pyrilamine; alkylamines, e.g., chlorpheniramine; pipera zines, e.g., hydroxyzine; and phenothiazines, e.g., promethazine. These drugs all have central nervous system effects in people. Second-generation antihistamines have fewer central effects. They include loratidine and terfenadine.

The involvement of IgE and histamine in equine airway disease is not yet clear. Investigators in Edinburgh have demonstrated an increase in histamine levels in BALF 4 h after stabling COPD-susceptible horses. However, antihistamines (H$_1$ antagonists) are generally of little use in the relief of airway obstruction in horses. Even if COPD is an IgE-mediated disease and histamine is released, the lack of efficacy of classical antihistamines is not surprising because many other bronchospastic mediators are also released during the inflammatory response. Serotonin

Serotonin (5-hydroxytryptamine, 5-HT) is included in this review because its concentration has been reported to increase in the plasma of horses with COPD. Platelets are a major source of both 5-HT and thromboxane. Thromboxane also is reported to increase in horses with heaves. In the lung, 5-HT facilitates the release of ACh from parasympathetic nerves and could therefore lead to bronchospasm. 5-HT exerts its effects by means of a plethora of receptors, and the importance of different subtypes in equine airways is unknown. However, 5-HT contracts smooth muscle in the trachea and small airways and increases the response of smooth muscle to activation of cholinergic nerves. There are no reports of the use of 5-HT antagonists for treatment of equine airway disease.

9. Eicosanoids

Eicosanoids is the term used to describe prostaglandins and leukotrienes, the derivatives of the metabolism of arachidonic acid, which is contained in membrane phospholipids. These compounds have a wide variety of actions in all organs, and the airways are no exception. The production of eicosanoids is initiated when phospholipase A$_2$ is activated either directly by a G-protein-coupled receptor or indirectly by an influx of Ca$^{2+}$ into the cell. The latter can arise in response to activation of a G-protein-coupled receptor or by physical disturbance of the cell membrane. Phospholipase A$_2$ releases arachidonate from the membrane phospholipids for subsequent conversion by means of either the cyclo-
oxygenase or lipoxygenase pathways into prostanoids or leukotrienes, respectively.

A. Prostanoids

Metabolism of arachidonate by means of the cyclooxygenase pathway yields the unstable prostaglandin H$_2$ (PGH$_2$), which is subsequently metabolized into a family of compounds including PGI$_2$ (prostacyclin), PGE, PGF$_{2\alpha}$, PGD$_2$, and thromboxane (TXA$_2$). There are two isoforms of cyclooxygenase, i.e., COX-1 and COX-2. COX-1 is constitutively expressed in all cells, but COX-2, which is induced by cytokines and other factors, is responsible for the increase in prostanoid synthesis that occurs with inflammation. Currently available nonsteroidal anti-inflammatory drugs (NSAID's) inhibit both COX-1 and COX-2. Inhibition of COX-2 is responsible for the beneficial effects of NSAID'S; inhibition of COX-1 explains the undesirable side effects such as ulcer production and renal damage.

The type of prostanoid produced by a tissue depends on the enzymes therein and their effects depend on the receptor subtype to which they bind and the coupling pathway that is induced. In equine airways, PGE$_2$ is produced by the mucosa, especially the epithelium. PGE$_2$ has high affinity for receptors that are coupled by means of G$_q$ to an increase in cAMP. For this reason, PGE$_2$ inhibits airway smooth muscle contraction and is generally anti-inflammatory i.e., it inhibits the release of other mediators. PGD$_2$ and PGF$_{2\alpha}$ have high affinity for receptors that are coupled to an increase in intracellular Ca$^{2+}$ and therefore contract airway smooth muscle and cause bronchospasm in most species.

In horses with heaves, there is apparently a shift in prostanoid synthesis from the anti-inflammatory compounds such as PGE$_2$ toward the proinflammatory compounds such as TXA$_2$. Despite this change in prostanoid profile, currently available NSAID's (e.g., flunixin) have no therapeutic benefit. This is probably because production of both proinflammatory and anti-inflammatory prostanoids is inhibited. It will be interesting to see if selective COX-2 inhibitors are more useful for the treatment of airway disease.

B. Leukotrienes

Leukotrienes are produced when arachidonate is metabolized by the 5-lipoxygenase (5-LO) enzyme. This is a cytosolic enzyme that is brought into proximity with the membrane-associated arachidonate by means of a 5-LO activating protein (FLAP). The production of leukotrienes can therefore be decreased by inhibition of either 5-LO or FLAP. Activation of 5-LO leads to the production of an unstable compound leukotriene A$_4$ (LTA$_4$), which is metabolized either to LTB$_4$ or LTC$_4$. The latter compound is further metabolized to LTD$_4$ and LTE$_4$. LTB$_4$ is a potent chemoattractant for neutrophils. The group of compounds LTC$_4$, LTD$_4$, and LTE$_4$ are collectively known as sulphidopeptide or cysteinyl leukotrienes and were formerly called slow-reacting substance of anaphylaxis. Because leukotrienes bind to receptors that are coupled by means of G$_q$ to an increase in intracellular Ca$^{2+}$, they are potent contracters of airway smooth muscle and mucus secretagogues. In the horse, LTD$_4$ consistently contracts peripheral airways but has a variable effect in the smooth muscle from the trachea.

Because leukotrienes are chemoattractive and cause bronchospasm and mucus secretion, there is great interest in modifying their production or blocking their receptors as treatment for airway disease. A FLAP antagonist blocks the early and late allergic responses in sheep airways and is now available for the treatment of asthma. It remains to be determined if similar compounds are useful in the treatment of airway disease in horses.

11. Corticosteroids

Heaves in horses, like asthma in people, is an inflammatory airway disease. Inflammation is associated with airway hyperresponsiveness to inhaled stimuli, mucus hypersecretion, airflow wall thinning, and bronchospasm. Suppression of the inflammatory response is therefore very effective therapy when the inciting environmental factor cannot be eliminated.

Glucocorticoids exert their beneficial effect in several ways. They diminish the number of circulating lymphocytes, inhibit the production of cytokines such as IL-2 that are at the top of the inflammatory cascade, increase the number of peripheral blood neutrophils by increasing release from bone marrow and diminishing removal from the circulation, diminish the production of eicosanoids by causing inhibition of phospholipase A$_2$, decrease the expression of leukocyte adhesion factors, and inhibit IgE-dependent histamine release. Corticosteroids also have important effects on $\beta$-adrenoceptors, which may explain in part the beneficial effects of steroids in airway disease. Corticosteroids increase $\beta$-adrenoceptor numbers and their coupling to adenyl cyclase so that the $\beta$-agonist-induced concentration of cAMP is increased in airway smooth muscle. Because many of the effects of corticosteroids involve effects on gene expression and protein synthesis, their effect is not immediate.

Corticosteroid administration is an effective treatment for equine COPD (heaves). Oral prednisone or prednisolone provide the most convenient treatment. They are administered at 1 mg/kg q 24 h for 1 week or until an improvement is observed, and then the dosage is decreased to the least that will prevent the clinical signs. Preferably this should be an alternate-day treatment. There are few side effects of long-term, low-dose prednisone or prednisolone treatment. More potent steroids can be used to initiate treatment of severe cases of COPD. One dose of triamcinolone (20-40 mg/500 kg IM) decreases airways obstruction for several weeks. Dexamethasone can also be used and is initially
administered at 0.1 mg/kg IV or IM for 2 days, and the dose is gradually reduced. Because prolonged use of triamcinolone or dexamethasone is likely to be associated with undesirable side effects such as lami- nitis, prednisone or prednisolone should be used for continued maintenance once obstruction is relieved.

In human asthma, use of inhaled corticosteroids has become standard treatment to relieve airway inflammation and obstruction.65 Beclomethasone dipropionate (Beclovent®), fluticasone propionate, budesonide, triamcinolone acetonide, and flunisolide (Aerobid®) are used by inhalation. This allows a high local concentration without the undesirable side effects of prolonged steroid administration. Asthmatic individuals titrate their steroid regime based on their own home-based measurements of peak airflow rates. Inhaled beclomethasone dipropionate is being used for treatment of horses with COPD.64 It is administered by means of a face mask (3500 µg/500 kg q 12 h). Improvement occurs within 4 days. After 2 weeks, treatment is administered once daily, and then the dose is gradually tapered to that necessary for maintenance. Anec- dotal reports indicate that a lesser dose (1000–1500 µg) is effective. It is not yet known if inhaled steroids cause adrenal suppression in the horse.

12. Cromolyn
Cromolyn sodium (disodium cromoglycate) and the similar compound nedocromil sodium are used prophylactically for the prevention of mild to moderate asthma. Their beneficial effect results from their ability to inhibit mast cell degranulation and therefore the release of histamine and the production of leukotrienes. Administration of cromolyn sodium (Intal®, 80 mg q 24 h for 4 days) to COPD-susceptible horses is reported to prevent the onset of clinical signs for up to 3 weeks following exposure to a dusty environment.66 Hare et al.67 have reported that cromolyn (200 mg q 12 h) was effective in treating small airway disease in a population of young race- horses with an elevated number of metachromatic cells (mast cells) in the bronchoalveolar lavage fluid. Cromolyn and nedocromil are available in metered-dose canisters and are administered by use of a face mask or spacer device. Because they prevent mediator release, these compounds must be used whenever a horse is likely to be exposed to the agents that induce airway obstruction.

13. Furosemide
In the late 1980's, Bianco et al.68,69 demonstrated that furosemide prevented exercise- and allergen-induced asthma in people. Subsequently, Broad- stone et al.70 demonstrated that furosemide given either intravenously (1.0 mg/kg) or by aerosol (1.0 mg/kg) relieved airway obstruction in ponies with COPD. The effect persists for several hours. The mechanism of furosemide's action involves the release of prostanoids and effects on neurotransmis- sion.71,72 The involvement of prostanoids was demonstrated by the prevention of furosemide's ef- fect in COPD horses by pretreatment with flunixin. There has been no study to determine the frequency with which furosemide should be used to treat airway obstruction in horses nor the effects of repeated dosing.

14. Aerosol Administration of Drugs to Horses
The treatment of airway disease in horses by use of inhaled medications has not been popular because the devices necessary to deliver the drugs were cumbersome, required the horse to wear a face mask for several minutes, and had low efficacy in delivering the medication into the lung. Several devices have now been developed for the more convenient delivery of aerosols with much greater efficiency.

The Aero-mask is a valved mask to which a spacer is attached.73 The horse wears the mask, and the drug is delivered from a metered-dose canister into the spacer from which it is inhaled by the horse. Use of this device does not require coordination between inhalation and activation of the canister. The drug remains in the spacer until the next inhalation. For the system to work most effectively, the mask must make a good seal with the horse face so that sufficient negative pressure is generated during inhalation to draw the medication from the spacer into the horse's lung. Because only one size of mask is available, the system does not fit tightly enough to be effective in all horses.

The spacer that is used in the Aero-mask can be purchased attached to a nose cone for use in hu- mans.1 This flexible cone fits over the nostril of most horses. The drug is delivered from a metered-dose canister into the spacer. The nose cone is applied to the horse's nostril and the drug is inhaled. We conducted a small trial of this system for delivery of pirbuterol to horses with COPD. It was effective in three of six horses. Given the low cost of this system, it may be worth a trial before buying the more expensive Aero-mask.

A handheld, metered dose, aerosol-delivery device has been custom built for horses and features a spacer that conforms to the horse's naris. The system efficiently deposits aerosol in the lung, and its use with pirbuterol results in consistent bronchodilation.36

With the advent of concern over the effects of aerosol propellants on the ozone layer, pharmaceuti- cal companies have sought ozone-friendly delivery systems. One of these uses a dry-powder inhaler. Drug is in capsules that are punctured in the inhaler device through which the patient inhales. This type of inhaler has been adapted to a closely fitting face mask for horses and has been used to deliver ipratropium bromide. It has the same disadvan- tage as the Aero-mask: Sufficient negative pres- sure has to be generated during inhalation to deliver the drug, and therefore the mask must fit tightly.

This research was supported by grants from 3M Animal Care Products and Bayer AG.
References and Footnotes


40. McKiernan BC, Koritz GD, Scott JS, et al. Plasma theophyll-


