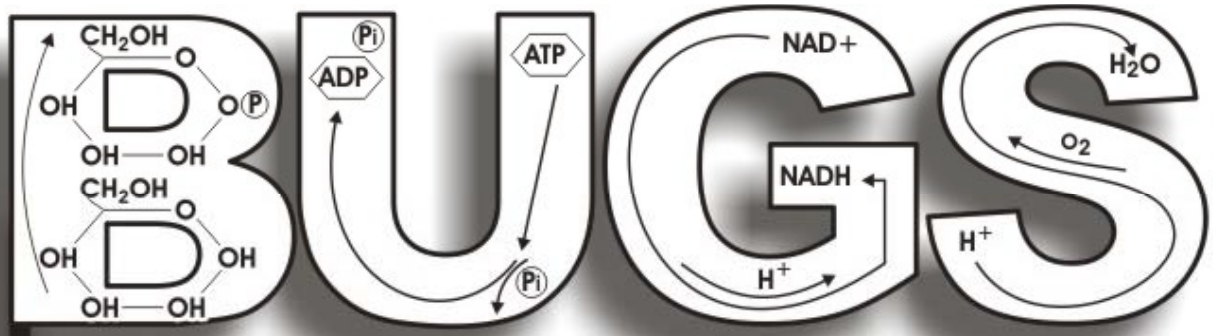
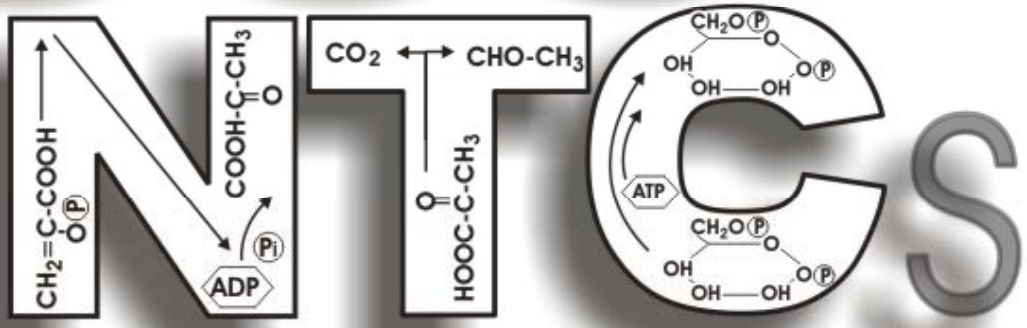




PHAR 301

SET #5 Lectures 12-13

ANNOUNCEMENTS

Tuesday, February 27th
&
Thursday, March 1st

The Princeton Review

MERCK FROSST

< Biochemistry Undergraduate Society - 5th Floor McIntyre rm. 511 >
< bugs@sus.mcgill.ca - (514) 398-5247>

Tuesday, February 27, 2007.
Dr Hales - Lecture #12

Asthma

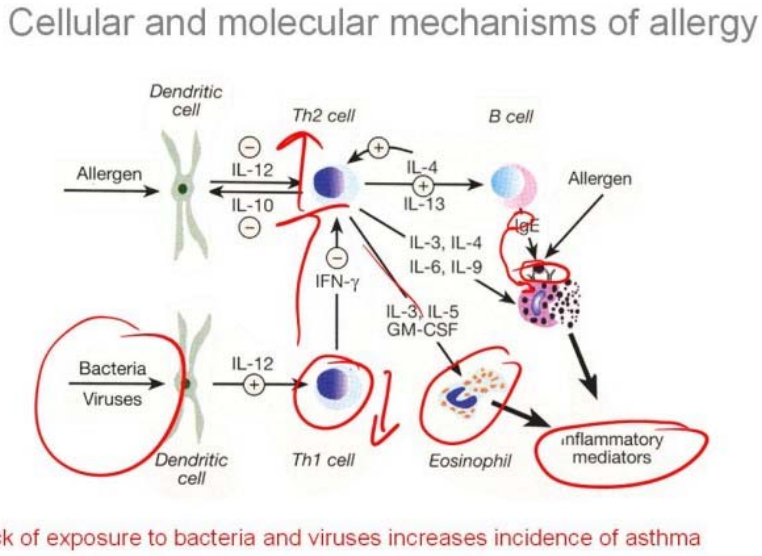
Today's lecture is covered in Chapter 34 of Brody

- Asthma is becoming more common in the society.
Two components to the disease:
1. Reversible airway obstruction; deal with it by broncho-dilation therapy.
2. Inflammatory disease (underlying cause of bronco-constriction); dealt with by anti-inflammatory therapy

- Asthma is very frequent in the western society. One out of four children have asthma in Britain and Australia.
- There are some genetics related to asthma but this is not the main cause.

- Asthma could be driven by the same mechanisms that drive allergy, but not all asthma cases have a clear cut allergen behind them.
So the cause of asthma is still a mystery.

Cellular and molecular mechanisms of allergy:



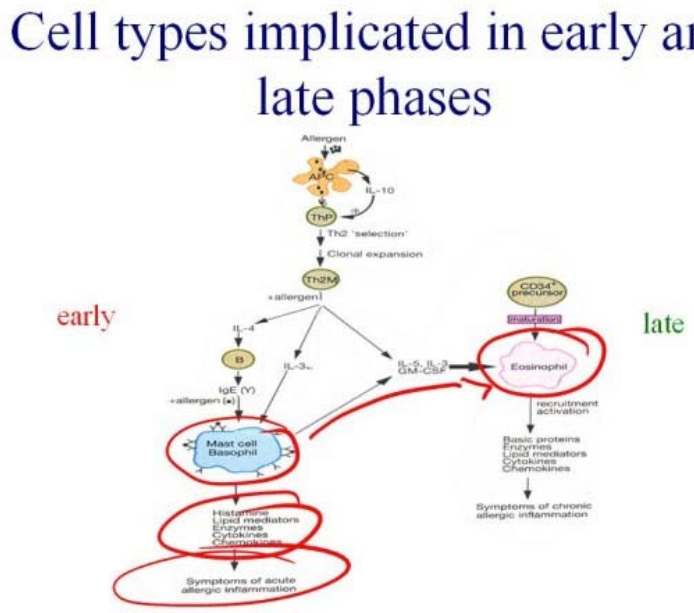
An allergen is presented by a dendritic cell to a T-helper type 2 cell (Th2). Th2 helps B cells develop immunity and proliferate. Th2 cells react with a B cell that has an immunoglobulin E receptor on its surface. IgE that are expressed on B cells are involved with allergy (ALL types of allergy, not only asthma. NOTE: not all asthma is caused by allergens, but a good component is) . IgE cells develop receptors which recognize the allergens. The IgE are hitting receptors for IgE on mast cells. Mast cells will then be degranulated and inflammatory mediators will be released.
Eosinophils are also important in maintaining hyper reactive bronchi. Th2 cells secrete cytokines which stimulate eosinophils.
T-helper type 1 cells (Th1) are designated to deal with bacteria and viruses.
There is a negative relationship between Th1 and Th2 cells: Th1 cells suppress Th2 cells through interferon gamma (IFN-γ). A higher level in Th1 cells will repress Th2 cells.

Hypothesis of why Th1 cells decrease and Th2 cells increase (possible causes of asthma):
1. The reason for increasing asthma incidence in our society is that we expose our babies to a smaller concentration of bacteria and viruses. Therefore, by trying to protect our children from bacteria, we create asthma.

2. Epigenetic factors are changes in how the genes are expressed and how the environment reacts with our genes. Environmental factors might change the gene expression and leads to an increase in Th2 and a reduction in Th1.

Asthma is usually connected with low social-economic regions. It happens to poor people in rich countries.

Cell types implicated in early and late phases:

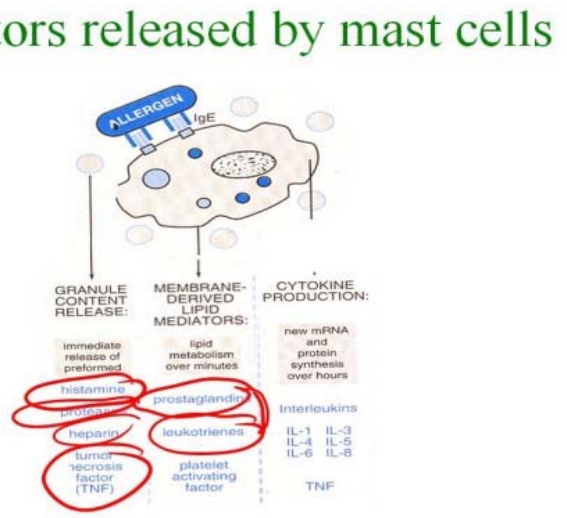


As explained earlier, an exposure by the allergen causes activation of IgE, which then signals for the degranulation of the sensitized mast cell. Mast cells release histamines, lipid mediators, enzymes cytokines and chemokines into the surrounding tissue causing several systemic effects and symptoms of acute allergic inflammation.

However, there is a longer impact driven by eosinophils. As the allergen activates mast cells, certain cytokines from the mast cells will activate eosinophils causing a later impact. The activation of eosinophils will cause the secretion of other basic proteins, enzymes, lipid mediators, cytokines and chemokines which will cause a chronic inflammatory response.

Mediators released by mast cells at the early phase include:

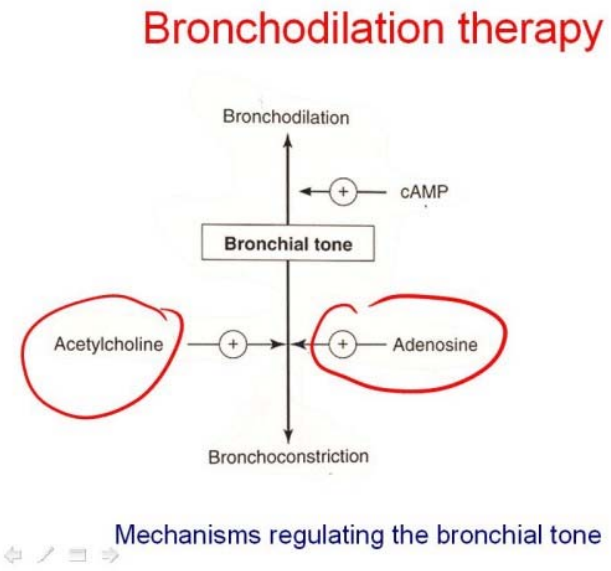
Types of mediators that mast cells release include:



1. Granule content release such as histamines, proteases, heparin, tumor necrosis factor (TNF).

   Note: Even though histamines are releases, antihistamines do not work in asthma.
2. Membrane derived lipid mediators such as prostaglandins, leukotrienes and platelet activating factor.
   - **Singulair**, a relatively new drug developed in Montreal inhibits leukotrienes.
3. Cytokine production of interleukines (IL-1, IL-3, IL-4, IL-5, IL-6 and IL-8) and TNF.

Bronchodilation Therapy :

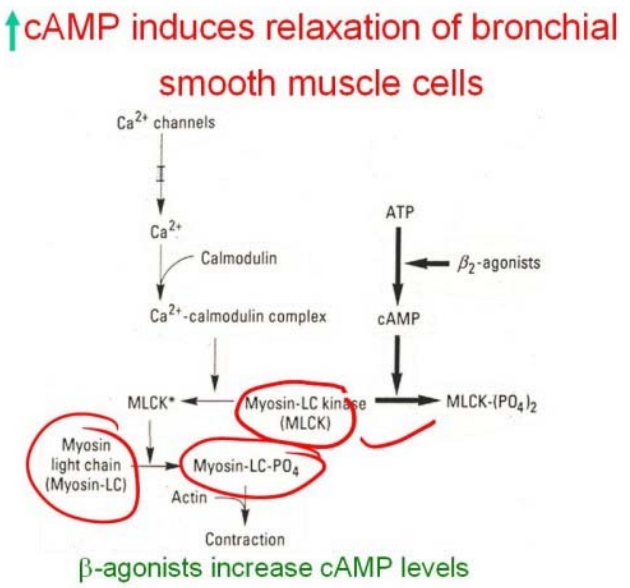


The bronchial tone keeps the bronchi on a regular stage, not totally dilated and not totally constricted. The sympathetic and parasympathetic systems regulate the bronchial tone. For example, the vagal nerve through acetylcholine and adenosine constricts the bronchi. On the other hand, an increase in cAMP through adrenergic receptors effects will dilate the bronchi.
Therefore, we can inhibit bronchoconstriction by two strategies:

- Inhibiting the action of acetylcholine and adenosine
- Stimulating cAMP.

First line of agents to cure bronchoconstriction:

1. Beta agonists that act on β-adrenergic receptors (G-coupled receptors that stimulate cAMP synthesis) could increase cAMP and cause bronchodilation.
2. Theophylline (form of caffeine founding tea) inhibits the enzyme phosphodiesterase which degrades cAMP. Therefore, as the degradation of cAMP is inhibited, its concentration increases. Moreover, theophylline is used to inhibit the adenosine receptor and that will inhibit bronchoconstriction and cause bronchodilation.
3. Muscarinic antagonists inhibit acetylcholine which inhibits bronchoconstriction and cause bronchodilation.

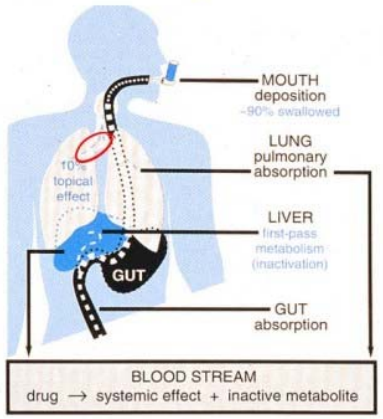


- Beta agonists increase cAMP levels. Constriction of smooth muscles involves an influx of calcium which binds to Calmodulin which activates Myosin-Light Chain Kinase (MLCK). The active form of MLCK activates Myosin-Light Chain (MLC) through phosphorylation. The phosphorylated form of MLC can interact with Actin to cause contraction. However, when the added beta-agonist activates cAMP, the Myosin-Light Chain Kinase (MLCK) gets phosphorylated and inactivated. The phosphorylated inactive form of MLCK will not activate MLC and as a consequence it will not cause contraction. (this is an important section for the exam)

The first class of drugs used against asthma is epinephrine. It is a beta agonist which increases cAMP and open bronchi, but it also increases the contraction of the heart muscles and that is not desired. Thus, selective beta agonist agents are used. They act selectively on the bronchi's beta-2 receptors and very poorly on the heart's beta-1 receptors.

- Episodes of increased asthma mortality accompanied the introduction of inhaled epinephrine (beta agonist) in the 1960s – but this was also associated with increased contraction of the heart (because there are adrenergic receptors in the heart)!

Systemic toxicity is reduced by using inhalers: increased topical versus systemic concentrations



Disposition of an inhaled drug

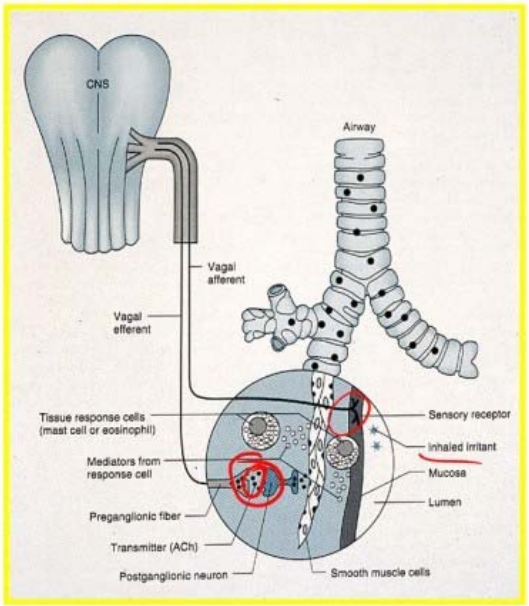
Selective β2 adrenergic agonistsact specifically on the bronchi smooth muscle and act very poorly on the β1 receptors found in your heart.
Selective β2 adrenergic agonists are used as nebulizers or inhalers to reduce systemic toxicity by increasing their topical concentration. 10% of the drug will act in the lung (bronchi) and increase cAMP levels there. The other 90% will be distributed to other organs such as the mouth, liver and gut. Examples of these inhalers include: Albuterol, Derbutaline, Perbuterol and Bitolterol.

Major problem with regular use of β2-adrenergic agonists:

One of the problems associated with β2 agonists used for a long time is exacerbated hyper responsiveness. The bronchi become more responsive to the agents that trigger asthma in that person. This is caused by tachyphylaxis which is using a β-adrenergic agonist or any drug agonist and the body responds by making that tissue hyperactive and so the person will need a greater dose of the drug to produce an effect. This is a feedback response. The cells will reduce the number of β-adrenergic receptors in an attempt to reach homeostasis.

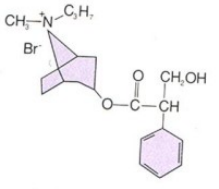
Long term administration does not reduce bronchial hyperresponsiveness.

Regulation of the bronchial tone by vagal firing

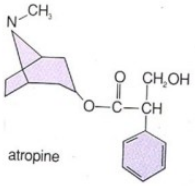


The vagus does the opposite - it causes constriction. The preganglionic fiber of the vagal nerve will secrete acetylcholine

Anticholinergic agents; atropine and nonabsorbable quaternary ammonium congeners iptratropium bromide



ipratropium bromide



atropine

(Ach). Ach acts on muscarinic receptors in the bronchi, reduces cAMP concentration and increases the contractility of the smooth muscle. Sensory receptors of the vagal nerve will sense the irritants, cold air, or other stimulants that will trigger asthma and will send this stimuli to the brain (central nervous system) and hit the ganglionic neuron. The ganglionic neuron secretes Ach, activates muscarinic receptors, reduces cAMP, and causes contraction.

Anticholinergic agents: (important to know the functions of atropine for the exam). Ipratropium bromide is the most commonly used as a bronchodilator.

Mechanism of action of Anticholinergic Agents:

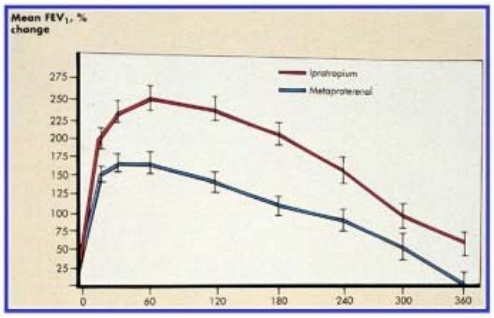
- The activation of M3 receptors results in the inhibition of adenine cyclase through the Gi (type of G coupled receptor that inhibits cAMP) and activatation K^+ channels.
    - Anticholinergic agents block these activities. Consequently, they increase cAMP. The increase in cAMP causes bronchodilation.

- However, anticholinergic agents aren't very effective drugs in asthma. They are effective in Chronic Obstructive Pulmonary Disease (COPD – usually precipitated by smoking).
    - The bronchodilation is less intense than the one produced by β2-adrenergic agonists.

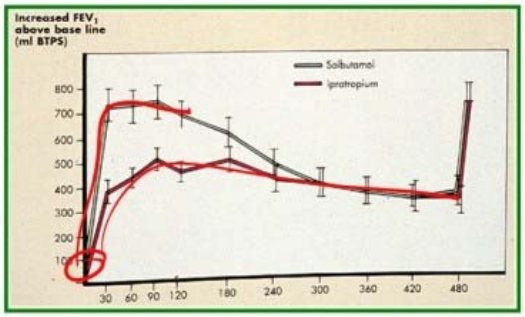
In asthma the cholinergic tone is not very important, so the anticholinergic agents will not have a very strong effect. In COPD the cholinergic input is very important and so anticholinergic agents will have a better effect than beta-agonists. The bronchodilation that a person will get with an anticholinergic agent in asthma will not be as powerful as the beta-agonist (therefore β-agonists are the mainstay for treatment of bronchoconstriction asthma).

Chronic obstructive pulmonary disease

COPD



Asthma



But, if we combine the two drugs: beta-agonist and iparatropium (anticholinergic agent), results in a greater and more prolonged bronchodilation because on one hand we increase the concentration of cAMP and on the other we block the reduction of cAMP.

- We use this combination if the person has a heart disease and asthma. Beta-agonists will still have an effect on the vasculature and the heart. To reduce the concentration of beta-agonists and reduce their effect on the heart, we add ipratropium.

To distinguish between an asthmatic person and a normal person, we measure the FEV (volume of air when you force exhale after inhaling). To evaluate the effectiveness of the drugs, we measure the increase in FEV.
- Solbutamol increases the FEV volume more significantly than ipratropium. Hence, in asthma the effect of a beta-agonist is much more powerful than an anticholinergic agent (muscarinic antagonist) so the muscarinic tone is less important. However, in COPD it is the opposite. It is much more important to reduce the vagal tone in CODP.

Methylxanthines: (another family of bronchodilators)

- Theophylline, a form of caffeine, exists in tea. Caffeine exists in coffee. (But need much higher doses than those found in tea to treat asthma!)

Mechanism of action:
- Methylxanthines inhibit phosphodiesterases at high doses.
- Phosphodiesterase degrades cAMP.
- Inhibiting phosphodiesterase increases cAMP.
- Also, methylxanthines can have an effect on the influx of calcium. Calcium is a villain agent involved in the contraction of the smooth muscles.
- Moreover, methylxanthines can cause membrane hyperpolarization, and uncouple calcium influx with contractility.
- But the most important function of methylxanthines is antagonism of adenosine receptors. Adenosine receptors play an important role in asthma by maintaining the bronchial tone. Adenosine receptors act to increase contractility but Theophylline and caffeine inhibit adenosine receptors and that inhibits contractility. Adenosine receptors are also G-coupled receptors which, in this case, cause reduction in cAMP. Therefore, if we inhibit these receptors we increase cAMP.

Briefly, it is important to remember that adenosine receptor antagonism is the most important effect of Theophylline and caffeine.

Positive and negative effects of methylxanthines:

Decrease drowsiness and fatigue
- Increase the flow of thoughts
- Reduces muscular coordination

At higher concentrations of caffeine and theophylline there is:
- Increase in CNS stimulation. The person becomes agitated and that can lead to :
  - nervousness and anxiety
  - increase in restlessness, insomnia and tremors

Higher toxic concentrations of caffeine and theophylline cause:
- increased focal and general convulsions
- increased activity of the medullary respiratory center which will lead to nausea and vomiting

Theophylline has a very **narrow therapeutic window**.

Cardiac effects of methylxanthines:
At very high concentrations they can cause tachycardia (increased heart beat) and arrhythmias.
- However, it has been shown that the risk of producing arrhythmia in normal subjects is low.

Diuretic effects of methylxanthines:
They increase the production of urine, increased defecation and relaxation. They increase the glomerular filtration rate and renal blood flow.

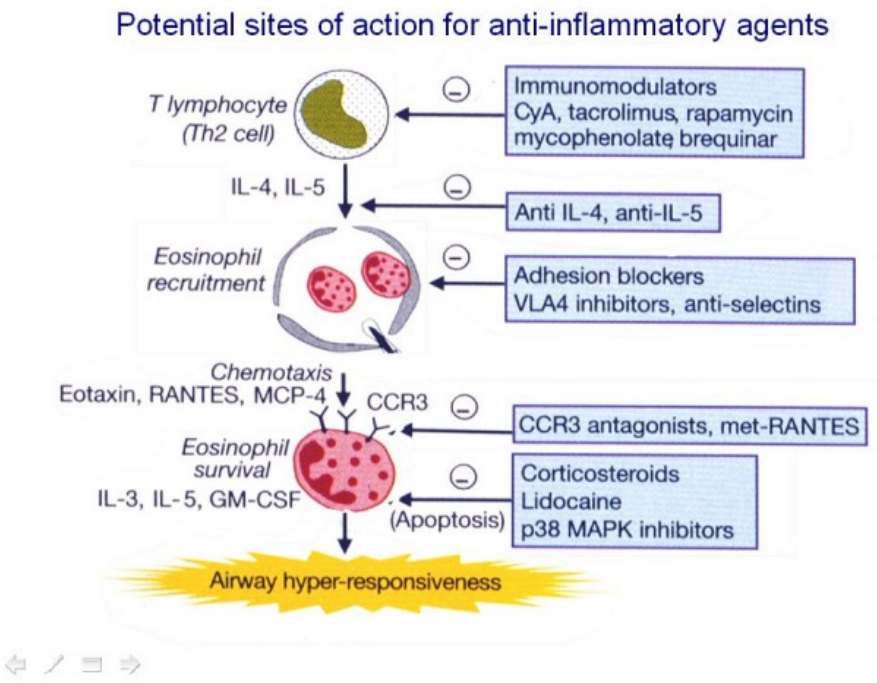
DNA antisense:
DNA antisense is a small sequence of DNA that is in the antisense orientation to your RNA. It is used as a drug for asthma. It is easy to make and is rational. It is a DNA sequence that fits the RNA sequence of the adenosine receptor (because want to knock down an adenosine receptor). It is very specific. However, it didn't come out yet.

Anti-inflammatory agents:

- Glucocortocoids
- Leukotriene synthesis inhibitors.

Mechanism of action:

Th2 cells recruit eosinophils and trigger the mast cells to eventually cause airway hyper responsiveness. Eosinophils have receptors like CCR3 and MCP4 that could be targets of anti-inflammatory agents.

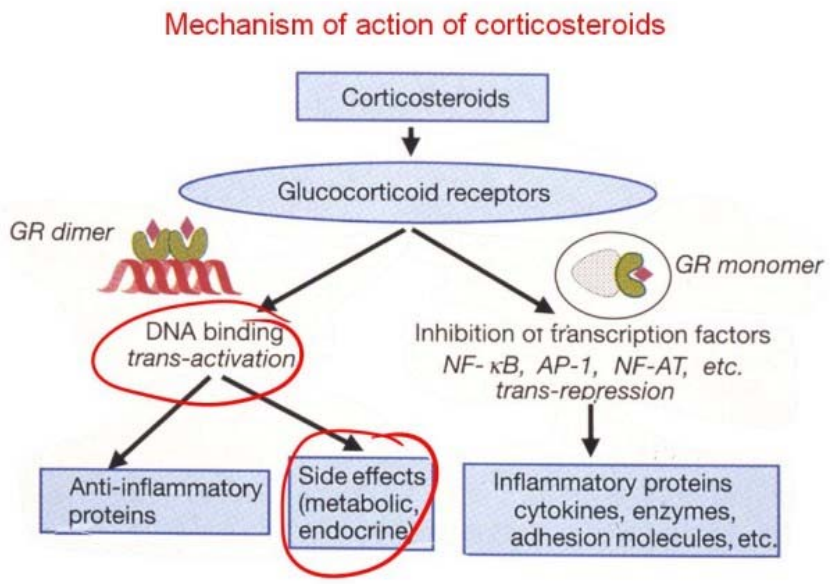


- Immunomodulators are used to inhibit T- cells (however risk that you are also inhibiting other T-cells).
- Anti-cytokines such as anti IL-4 and anti IL-5
- Adhesion blockers to block the recruitment of eosinophils
- Antagonists against CCR3 receptors
- Corticosteroids inhibit the recruitment of eosinophils by blocking cytokines.

Mechanism of action of corticosteroids:

Corticosteroids reduce the inflammation that precipitates the constriction. They bind to the glucocorticoid receptors. Glucocorticoid receptors are receptors of the nuclear receptor family which are either in the nuclei or going to the nucleus upon the interaction with the ligand. Then, they find sequences in DNA and bind to them. They essentially act as trans-activators. Conversely, they can recognize genes and recruit repressing machinery that will repress the gene expression. Hence, glucocorticoids can activate or repress gene expression.

- Corticosteroids have anti-inflammatory activities by either stimulating anti-inflammatory agents or reducing inflammatory proteins. They also have metabolic and endocrine side effects.



-In addition, corticosteroids are responsible for recruiting glucose, so therefore they will breakdown bone, muscle and fat to make glucose.
- They were created for our stress response. They are one of the most powerful agents that are stimulated upon stress. So essentially, glucocorticoids are destructive agents whose purpose is to answer an acute need for energy to escape a threat.
- Another kind of damage that glucocorticoids do is to fool the adrenal into thinking that the adrenal does not need to produce glucocorticoids and therefore you get hypertrophy of the adrenal.
As indicated previously, glucocorticoids can act by inhibiting transcription factors and prevent them from accessing their recognition sites. As a consequence, it inhibits cytokines and enzymes, inflammatory proteins, adhesion molecules, etc…

To reduce the toxicity of glucocorticoids, we use aerosol.

Note: Glutocorticoids are DANGEROUS! But sometimes essential!
Glucocortocoid therapy is used for subjects who require β2-adrenergic agonists more than 4 times a day. In this case **belcomethasone**, an inhaled form of glucocorticoids, is given 4 times a day - it has a very limited toxicity.
To reduce the hyper responsiveness, we keep those patients on maintenance.

Remember: Glucocorticoids are not going to work if you can't breathe – they solve the basic inflammatory response but they are not dealing with the acute situation which is 'pipes' constricting. Rather, what they can do is prevent the inflammation that precipitates the concentration. Therefore the way to figure out whether the glucocorticoids work is to ask the patient how often they need to use the beta-agonist puffer.
Systemic glucocorticoids (injection) are used for acute exacerbations and chronic severe asthma (patients take glucocorticoid pills for years to keep it under control!).

- When patient comes to clinic with severe asthmatic attack, you immediately inject the patient with β-agonist (puffer is not enough) and at the same time you inject glucocorticoids.

To resume, there are 3 stages of therapy:

1. For very mild asthma of few attacks, we use beta-agonist puffers.
2. When we have to use these puffers more frequently, we use puffers of belcomethasone or glucocorticoids.
3. When the case gets severe, we use systemic glucocorticoids.

NSAIDS (nonsteroidal anti-inflammatory agents):

NSAIDS do not have adverse effects as glucocorticoids do. But they are not used for asthma therapy because they aggravate asthma by stimulating leukotrienes synthesis. As the synthesis of archidonic acid is activated, it could synthesize prostaglandins (PGs) by the enzyme cyclooxygenase and leukotrienes. NSAIDS inhibit the enzyme cyclooxygenase and the production of PGs. As a result, the production of leukotrienes is triggered more and more leukotrienes are created and that aggravates asthma.

Leukotriene inhibitors:

- Zafirlukast is an LTD4 receptor antagonist - inhibits LTD4
- **Zileuton** is an inhibitor of the enzyme 5-lipoxygenase.

Mechanism of action of Leukotriene inhibitors:

Leukotrienes are involved in bronchoconstriction, bronchial reactivity, mucosal edema, and mucus hypersecretion and these are all involved in asthma!
Therapeutic advantages of leukotriene inhibitors:

- They can be taken orally
- They are approved for young children
- Montelukast are used only once daily - **Singulair**.

The last group of drugs are:

Mast cell-stabilizing agents:

The degranulation of mast cells triggers both the early phase and the late phase of asthma.
Cromolyn Sodium is the family of mast cell stabilizing agents. There are different analogues of it.

Mechanism of action:

- Mast cell stabilizing agents inhibit pulmonary mast cell degranulation and asthma attack.
- They reduce the release of leukotrienes and histamines.
- They also can reverse the functional changes induced in leukocytes in response to allergens.
- They inhibit the activating effect of chemoattractant peptides on neutrophils, eosinophils and monocytes.
- They do not have a bronchodilating capacity; they don't work on the bronchi (like glucocorticoids!) – therefore, used to PREVENT.
- They reduce the calcium influx.

Thursday, March 1st, 2007
Dr Clarke - Lecture # 13

Colds, Allergies, Anti-histamines, Cold Remedies

Drugs and terminology to know by name:

- Phenylpropanolamine (PPA)
- Phenylephrine
- Pseudo(ephedrine)
- Diphenhydramine (first-generation, sedating antihistamine)
- terfenadine (non-sedating antihistamine)
- Sympathomimetic
- Mucosa
- Edema
- Perennial Rhinitis, rhinorrheoa

Sections to read in the book:

- Chapter 10 – Drugs affecting the sympathetic nervous system
  - Whole chapter is useful **revision** – but most people probably won't need to read Brody for this lecture
  - Only page 123 is directly relevant to sympathomimetics
- Chapter 54 – Histamine and antihistamines
  - Read only page 701 and H1 antagonists section to end of chapter (pg 707-709)

The Nose

- Erectile tissue
- Various functions, one is to warm and moisturize the air coming in from the outside
- **rhinitis** = inflammation of the mucosa (lining), characterized by:
  - ✓ **vasodilation** - increased blood supply
  - ✓ **edema** - cells of the capillaries contract creating gaps in capillary wall through which fluid enters the tissue and causes swelling
  - ✓ **mucus secretion (rhinorrhea)**
- Blocked/stuffy nose = mucosa membrane/lining (spongy tissue) in enclosed space becomes inflamed – can only expand inwards because of bony cavity of the nose
  - Blood vessels become inflamed and the tissue becomes edematose
  - If you also have mucus secretion, then you have a **runny** nose

Three ways to deal with a stuffy nose

1. **Sympathomimetic –** mimics the sympathetic nervous system (adrenaline or noradrenaline)
2. **Antihistamine** – works if the stuffy nose is the result of an allergic reaction
3. **Stick a hose-pipe up your nose –** not recommended! (don't need to know)

- For a cold, your best bet is a nasal decongestant while as for hay fever, both nasal decongestants and antihistamines can be effective

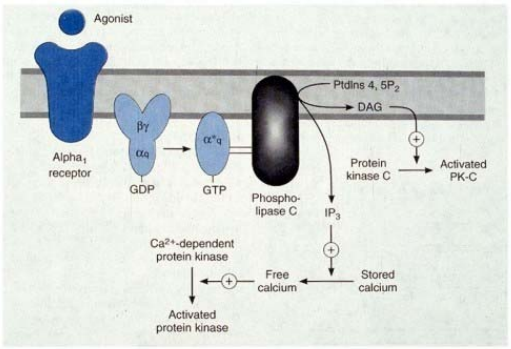
1. SYMPATHOMIMETICS

- The blood vessels supplying the nose have α-1 and α-2 adrenergic receptors, which produce vasoconstriction when stimulated.
- When you have a stuffy nose you have blood vessels that are dilated, therefore you want to target one or both of these receptors to produce vasoconstriction. Preferably both receptors.
- Noradrenergic terminal (picture): axon, a terminal and an axon coming out the other end of the terminal
- Some nerve terminals end on a branch of the axon, others are not really terminals but are more like beads on a string – could be thousands of such "terminals" on one axon
- **Main point:** at a noradrenaline terminal, noradrenaline is released. Some drugs enhance the release of noradrenaline from the terminal (indirectly acting – ex. pseudoephedrine, PPA), while others act directly on the post-synaptic α-receptor (ex. phenylephrine).

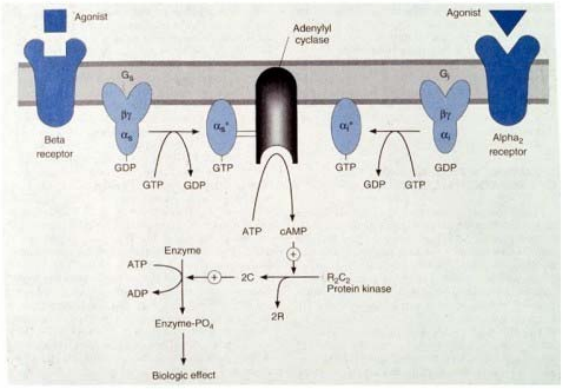
Mechanisms

- stimulation of **α-1 receptor** results in an increase in diacylglycerol (DAG) and IP3 (second messengers) which in turn produce an increase in activity of **PKC** (protein kinase C) and free calcium in the cytosol; results is **vasoconstriction** indirectly

α1 receptor stimulation



α2 receptor stimulation



- **α-2 receptors** work through G-protein coupling inhibit **adenylate cyclase**, decreasing **cAMP** levels, which in turn reduces the activity of **PKA** (protein kinase A); end result is **vasoconstriction**

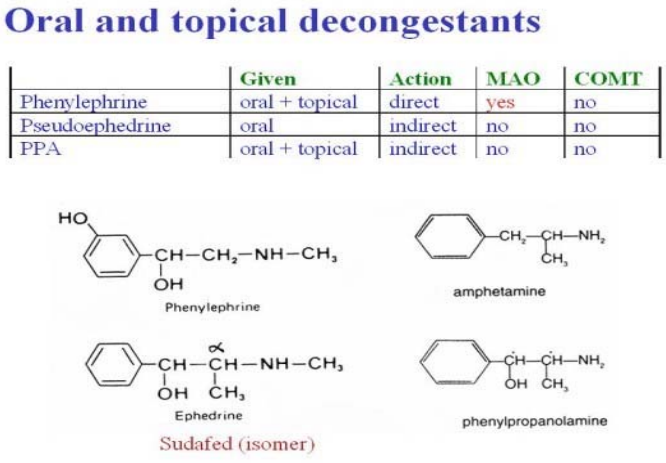
Ephedra (Ma Huang)

- Chinese Herbal Remedy to treat asthmatic wheezing
- Was known for many years that if you take this plant when you have asthma it will loosen up your bronchi and you'll feel better
- 1920's – asthmatics couldn't take adrenaline orally because it has poor oral availability (not well absorbed in the gut and is broken down in the blood stream); unless one had access to a doctor who could inject a high dose of adrenaline intravenously, it was of no use
- Needed something that could be taken orally and didn't require injection
- K.K. Chan – Chinese medicinal chemist in the USA; extracted the active ingredient from the herbal remedy; was found to be **ephedrine**
    - Only bad thing was that it was not fast-acting because it was an oral drug

- Became unavailable because of war between communists and other forces in China

➢ 1930's - Drug developers made **amphetamine (Benzedrine)**; fine powder that could be volatilized and inhaled (enters blood stream quickly), became very popular OTC drug (used as decongestant, asthma, and even for students preparing for exams!), even offered on airlines; eventually realized that it could become addictive

Oral and Topical decongestants



✓ Mimics noradrenaline
✓ Noradrenaline is a catecholamine that is broken down by **catecholamine methyltransferase** (COMT, enzyme widely distributed in body) and monoamine oxidase
✓ However, the drugs that follow are not catecholamines and hence are not broken down by COMT
✓ Another drug that breaks down noradrenaline rapidly is **MAO** – as we will see, phenylephrine is broken down by MAO
✓ As a general rule, these drugs are longer lasting than adrenaline

1. **Ephedrine** – buy as Sudafed (aka. pseudoephedrine, stereoisomer of ephedrine, since it contains two asymmetric carbons); **indirect agonist –** forces the release of adrenaline
   - can be used to make methamphetamine (crystal meth) – Sudafed is the precursor to make methamphetamine
2. **PPA –** off the market now because it caused strokes in patients; **indirect agonist**
3. **Phenylephrine – direct agonist**, acts directly on the receptor; broken down by MAO, therefore has no effect when given orally

Ferret - used as animal model to test nasal congestion
Analogous to **Ohm's Law** – to determine resistance, you apply pressure and measure the airflow; useful in testing new drugs as decongestant, but you need to anesthetize the ferret first, therefore there may be drug interactions

Experiment:

➢ Ferrets were given the cold virus and then split into 2 groups (control was given placebo and became congested, the others were given a decongestant and got better)
➢ **Question:** The drug group has rebound resistance and their nose was stuffing up after the drug was stopped. Why?
➢ **Rebound Effect:** the drug is pushing against the body in one direction so the body compensates (acute tolerance), when the drug is quickly removed the body is pushing in the opposite direction and you get a rebound effect (ex. Stuffy nose)
➢ **Remember, these drugs will not cure a cold, they will simply relieve the symptoms**

Most of this research is done in **conscious humans**

➢ At any given moment one nostril is more congested than the other; alternates every 2-4 hours between the nostrils (cyclical)

Experiment:

- **Placebo** – no effect
- **Phenylephrine** (direct agonist) – administered as a nasal spray; has a big effect; maximally active within 15 minutes
    - If taken orally though (still sold in this form) ineffective because broken down by MAO
- **PPA** (indirect agonist) – syrup, orally administered; takes a long time to have an effect and long time for the effect to disappear; presumably slow absorption in the gut
- **Ephedrine** (indirect agonist) – inhalation; very rapid effect; wears of very quickly

❖ **From this, you cannot tell whether the differences in the action of the drugs are due to the properties of the drugs themselves or due to the route of administration. Likely it is a combination of both.**

Effects on Blood Pressure

- Since sympathomimetic drugs cause vasoconstriction then they may also have an effect on blood pressure (increasing it)

PPA – causes increase in blood pressure (banned because this property caused hemorrhagic strokes (brain) and heart attacks in some patients)

- By stimulating noradrenaline/adrenaline release, PPA will produce β1 receptor stimulation in the heart, which results in increased heart rate and stroke volume and in turn, increased blood pressure
- **Is there another mechanism by which PPA increases blood pressure**? Yes.
- α-1 receptors are widely distributed, including smooth muscle on blood vessels; therefore, stimulation of α-1 receptors in the smooth muscle of blood vessels causes vasoconstriction increasing total peripheral resistance which results in increased blood pressure

Phenylephrine – DOES NOT cause increase in blood pressure

- this is likely because it is broken down by MAO
- If some phenylephrine did enter the blood stream it would act on the α-1 receptors in the blood vessels (not the β receptors in the heart since it is a direct agonist) and should cause vasoconstriction and increased blood pressure. However, increased blood pressure is not observed. Two possible explanations:

1. Not around in high enough concentration because it is broken down by MAO.
2. Since phenylephrine does not act on the heart β1 receptors, the vasoconstriction may be countered by reflex bradycardia (slowing of the heart rate), which would prevent an increase in blood pressure

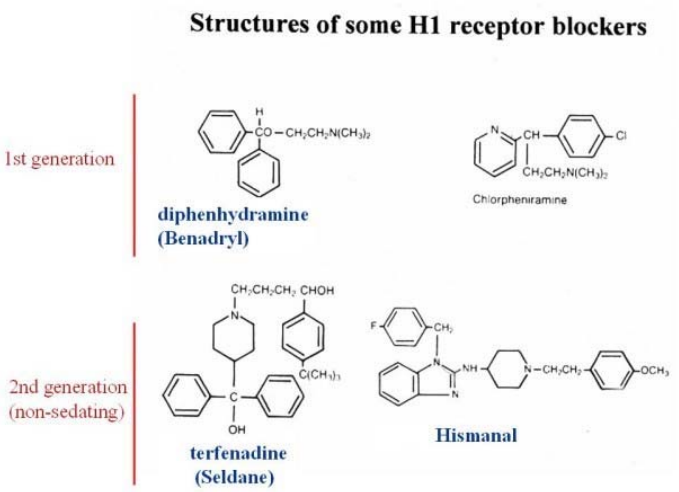
2. ANTIHISTAMINES AND ALLERGIES

- Seasonal Allergic Rhinitis – **Hay Fever** - Not related to hay, nor is it a fever (ex. Ragweed)
- Perennial Allergic Rhinitis (throughout the year) – cat, dust
    - Generally speaking, we give the same treatment for both types of rhinitis

Allergic Reaction and Antihistamines

- There is an immune response. Antibodies recognize and attach themselves to the allergen (ex. pollen) sensitizing the immune system; they then attach to mast cells which release inflammatory mediators, including histamine); histamine acts on the H1 receptor subtypes
- **NOTE: Anti-histamines in this lecture refers to antagonists of H1 receptors. There is another class of H2 receptors that are blocked by other drugs and are involved in ulcers.**

- Anti-histamines only act against histamine, but do not interfere with the other inflammatory mediators, which is why they are not fully effective (about 30% of people don't respond adequately to anti-histamines).
- 2 classes of anti-histamines: First generation and Second generation
- **First Generation:** the only thing available at the time for Hay Fever; gets in the brain (crosses BBB) and makes you very drowsy!
- **Second Generation:** more recent; DO NOT get in the brain (can't cross BBB) so they don't make you drowsy!



Anti-histamines and the Common Cold

- **Anti-histamines cannot treat the common cold!**
- Why? Because histamine release does not increase in the common cold
- However, bradykinin (another inflammatory mediator) release does increase, therefore a bradykinin antagonist might be effective in treating the cold
- Even though they don't work, anti-histamines are still found in some cold medication and sleeping pills, which could be dangerous as a study showed an increased incidence of car accidents among people taking antihistamines
- However, remember that **correlation does not necessarily imply causation!**

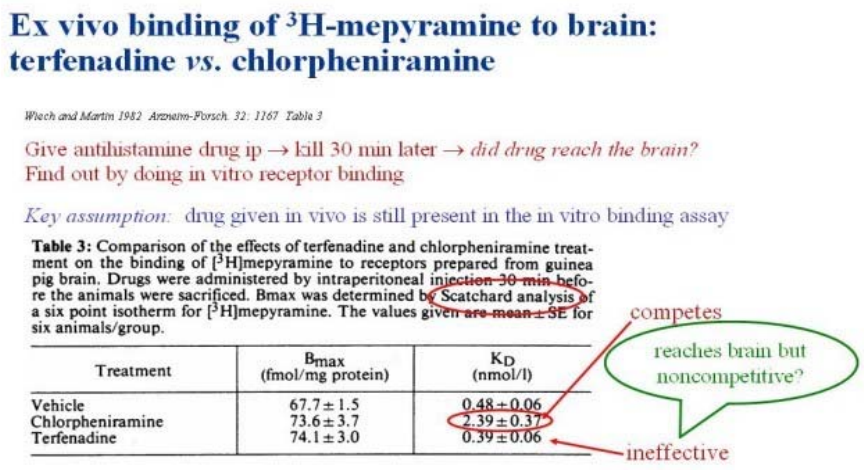
terfenadine (Seldane)

- The first Second Generation, non-drowsy antihistamine was developed deliberately so it could not cross the BBB and get into the brain; it was called terfenadine (Seldane™) is no longer sold
- Made lots of compounds, checked if they blocked H1 receptors and then tested whether or not the got into the brain
- To test if a drug crosses the BBB, you can give a radioactive form of the drug systemically (IV) and then see how much gets into the brain or spinal cord
- **Autoradiography**: inject the animal with the radioactive compound, kill it, freeze it, cut it into thin sections, expose each slide to x-ray film and see where the radioactivity is present.
- When the drug does not enter the brain much, over time it accumulates in the GI tract from where it is excreted

- **Two** limitations to this approach:
  1. It might have been broken down in the brain - perhaps it isn't radioactive terfenadine showing on the film (experiment measures radioactivity) but rather a metabolite of it.
  2. We don't know how dark the film needs to be in order for there to be a pharmacologically important effect. It is possible that you may need only minute amounts that aren't detectable by the film because it is not sensitive enough.

- **How much of the drug can get in the brain before its pharmacological effects on the brain are observed?**

- To test: give the drug in vivo, kill the animal, take the brain and do a binding assay to see whether the radioligand (ex. Radio labeled histamine) binds the H1 receptors. Have to assume that if the drug had reached the brain it would still be present in the binding assay enough to inhibit the binding of the radioligand. (Scatchard Analysis: used to determine the affinity and density of the binding sites for the drug that binds to H1 receptors)



- **Positive control:** Clorpheniramine, an H1 **competitive antagonist** that was known to get into the brain. Animals that had received it showed a change in binding affinity of the radioligand. Therefore the assumption was valid.
- Mice treated with terfenadine did not show a change in affinity for the radioligand. Thus it appeared that terfenadine didn't enter the brain and had no effect on the H1 receptors in the brain.

Have to consider:

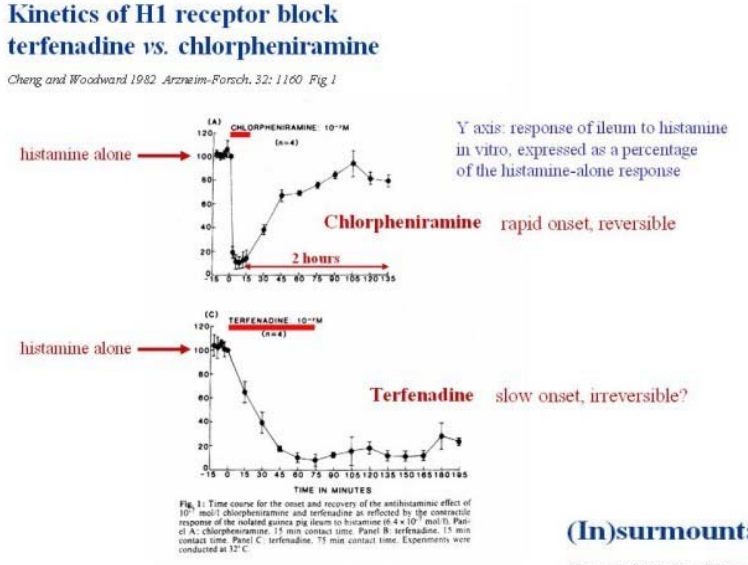
1. The possibility that terfenadine does in fact enter the brain and act on H1 receptors, but in a **non-competitive manner**, binding the receptor at a different site than the radioligand. **Found not to be the case.**

2. One possible explanation for the lack of terfenadine effect in the brain is that the dose of terfenadine was simply insufficient. In fact, maybe the amount of terfenadine given was not enough to occupy H1 receptors even in the periphery; tested this idea by doing a comparable study but instead, with H1 receptors from the ileum (GI tract). Found that the dose of terfenadine given in the previous study was enough to occupy about 90% of the peripheral H1 receptors (the control drug, chlorpheniramine, also occupied approximately 90% of H1 receptors in the ileum) – therefore, this proves that terfenadine was given in a high enough dose to occupy brain receptors if it passes freely across the blood brain barrier.

3. We also have to consider the possibility that brain H1 receptors could be less sensitive to terfenadine than H1 receptors in the periphery. To test this hypothesis, experimenters took brain tissue and **directly** applied the radioligand in vitro to see whether terfenadine would actually interfere with the binding. They found that terfenadine inhibited the radioligand binding to the same extent as in the ileum, therefore this shows histamine receptors in the brain are NOT less sensitive to terfenadine, as long as terfenadine reaches its target.

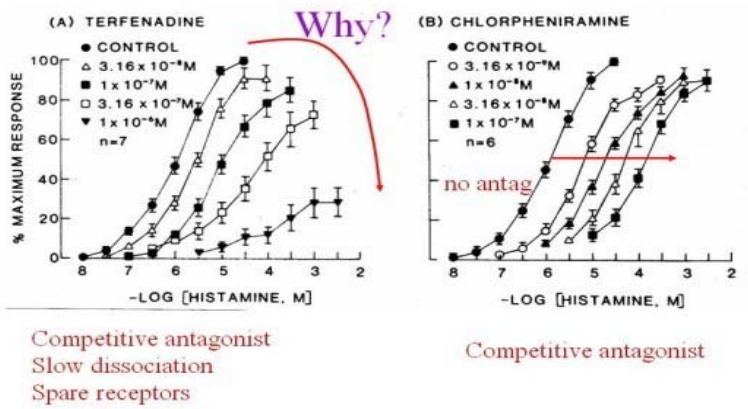
- **Final experiment**: simultaneously inject the radioligand intravenously in mice at the tail and give test drugs (all by the same route) and observe the amount of radioligand binding.
- The drugs that inhibited the binding of the radioligand to the H1 receptors in the brain were all First Generation drugs, while the drugs that did not significantly inhibit the binding of the radioligand to the H1 receptor in the brain were all Second Generation drugs. Thus, **Second Generation drugs do not cause drowsiness because they do not enter the brain.**

Other pharmacological principles

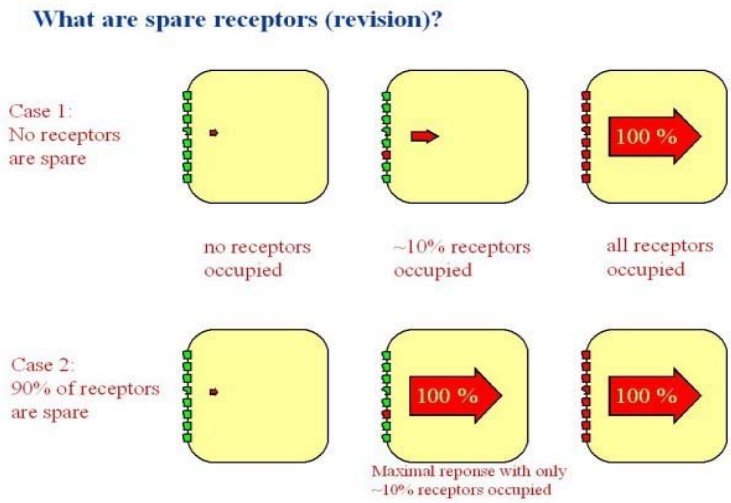


- Terfenadine produces a large blocking effect -however it takes a long time before the drug reaches its maximal effect (1 hour, as opposed to 1st generation drug chlorpheniramine, which has a rapid onset). Also, once the drug is discontinued, that which is already bound does not come off (irreversible antagonist) within the timeframe of the experiment (i.e. several hours).

- **Chlorpheniramine** – increasing concentration of the drug chlorpheniramine causes the dose-response curve for histamine to shift to the right; the maximal effect of histamine can be achieved but at higher concentrations; classic behavior of a competitive reversible antagonist
- **Terfenadine** – at low doses of terfenadine the dose-response curve for histamine shifts to the right, but high concentrations of terfenadine pull the curve down; however, we know from previous experiments that it is a COMPETITIVE antagonist (therefore this downward effect in not due to noncompetitive inhibition)



- *Why does terfenadine produce results typical of a non-competitive antagonist when it is known to be a competitive antagonist?* Answer: **Spare Receptors combined with "irreversible" (actually very slow) association.**



- When there are no spare receptors, in order to get a maximal response ALL of the receptors must be occupied by the agonist. If only 10% of the receptors are occupied there will be a minimal response (e.g. 10% of the maximum possible response).
- When there are spare receptors, only a fraction of the receptors need to be bound to produce a maximal response. So, even if only 10% (say) of the receptors are occupied by the ligand one may still get max response. If more agonist is given and more than 10% of the receptors are bound the response will not increase further.
- At the molecular level, spare receptors are not different from the ones that are actually occupied, which ones are occupied at a given moment is based purely on chance.
- Therefore, at low concentrations, terfenadine binds the receptor, but the ligand can still bind to other spare receptors to produce a response; at high concentrations terfenadine will irreversibly occupy most of the receptors (ex. 95%) and there will not be enough spare receptors left for the ligand (histamine) to bind to produce the maximal response.

Sedation and Effects on Driving

- Some people given antihistamines (diphenhydramine (1st Generation) **or** terfenadine (2nd Generation), others given placebo (control)
- Put in a car with a driving instructor and a technician in the back who is measuring the distance of the car from the side of the road (ability to keep the car a fixed distance from side of the road) – done under double-blind studies

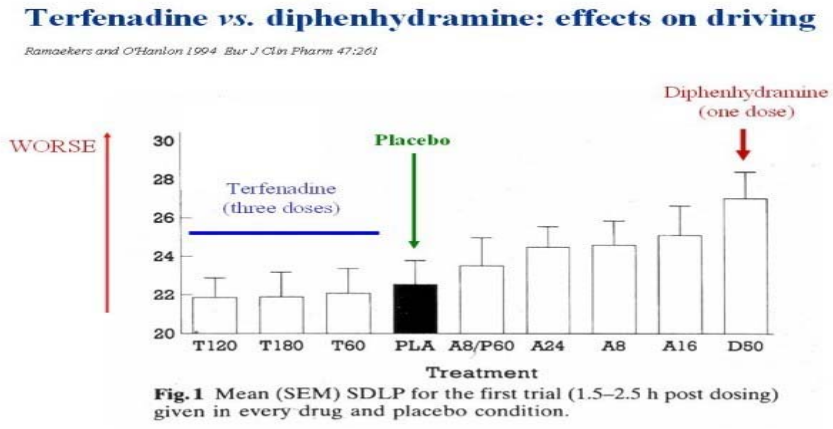


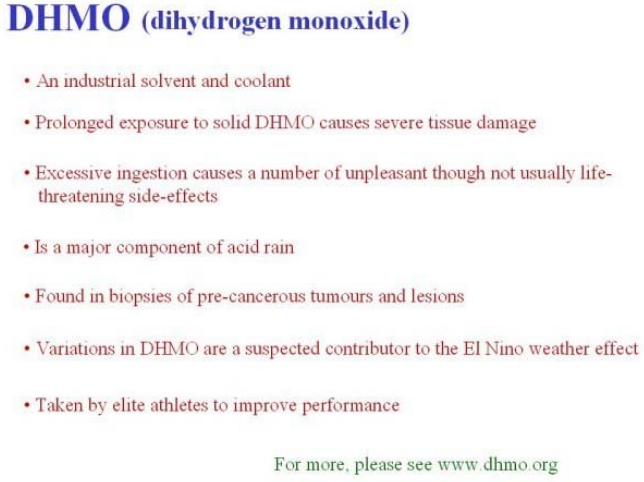
Fig. 1 Mean (SEM) SDLP for the first trial (1.5–2.5 h post dosing) given in every drug and placebo condition.

- main outcome in this study is the STANDARD DEVIATION of the distance from the side of the road
- terfenadine had similar standard deviation to placebo ; diphenhydramine had a significantly higher standard deviation (therefore more weaving of the car from the side of the road)

- **Older antihistamines are just as good as newer antihistamines at relieving symptoms, the newer ones simply have fewer side effects, e.g. no drowsiness**

Combining anti-histamines with nasal decongestants

- In a study patients with Hay Fever received either an anti-histamine alone, Sudafed alone, or a combination
- The beneficial effects were mostly due to the nasal decongestant (ex. Sudafed) – odd results
- General conclusion from observation of many studies: for allergic rhinitis, nasal decongestants are good, antihistamines are good and your best bet is to take a combination of both



Side Note: Dihydrogen monoxide (DHMO) is an environmental pollutant – read up about it! http://www.dhmo.org/ This might be an exam question!!!

Terfenadine

- Gets converted to an active metabolite very quickly – the active metabolite is the active component that is responsible for the H1 antagonistic effects of terfenadine
- Certain drugs (ex. antifungal drugs) or grapefruit juice inhibit the P450 enzyme that convert terfenadine to the active metabolite
- This results in a build up of terfenadine, which has adverse effects on the heart and causes a potentially fatal ventricular arrhythmia; taken off the market
- Once terfenadine was taken off market, the active metabolite was turned into a drug = Allegra
- **Allegra** – very safe, avoid the heart problems because it is downstream of that

- Some side effects of the First Generation H1 antagonist are beneficial, others are adverse. Many drugs contain H1 antagonists to make use of their beneficial side effects
- Histamine receptors are G-protein coupled receptors with 7 transmembrane domains – molecularly related to muscarinic and adrenergic receptors; therefore some H1 antagonists also act to some extent on adrenergic and muscarinic receptors – accounts for side effects

Side Effects of H1 Antagonists

1. **Muscarinic receptor block** – dry mouth, alleviates motion sickness
2. **Sodium Channel Block -** some people are allergic to lidocaine (local anesthetic); there is an H1 antagonist that happens to block sodium channels by chance that can be used as a local anesthetic instead. This is useful in patients that are allergic to the standard sodium channel blocker local anaesthetics.
3. **α-1 Receptor Block** – H1 antagonists may act on α-1 receptors in the smooth muscle of blood vessels to cause postural hypotension; feel faint upon standing up
4. **Drug-drug interactions** – via competition for hepatic P450 enzymes
5. **Allergy** – may be allergic to the anti-histamine

6. **Sedation** – First generation anti-histamines cross BBB and block H1 receptors in the brain; commonly found in sleeping pills that are available to everyone

H1 receptor blockers in OTC medications:

7. **Motion sickness** (Dramamine, Bonine, Marezine)
8. **Cold/Flu (Day/night pills)** – morning pill is a sympathomimetic (nasal decongestant) doesn't make you drowsy; the evening pill has an H1 blocker that makes you drowsy and helps you go to sleep

Glucocorticoids

- More effective for treating allergies than antihistamines
- Taken as nasal inhaler – not as quick acting as antihistamines, but if taken on daily basis during ragweed season, they are more effective for the average patient
- Do not act immediately, slow offset

Treatment of Cold Symptoms

- Symptoms: Increased blood supply, edema, mucous secretion
- Muscarinic receptors – produce dry mouth BUT can also **decrease mucous secretion (rhinorrhea)**

Summary

- Stuffy nose due to: colds **OR** allergies – each require different medication
- Allergies involve histamine release, colds do not.
- Allergic histamine release can be blocked using anti-histamines, but allergies also involve the release of other inflammatory mediators that are not blocked which is why antihistamines are not totally effective
- Colds do not involve consistent release of histamine, therefore antihistamines work very poorly or not at all
- The route of administration is critical – can determine the rate of onset and offset of a drug effect
- If you have a runny nose, you would think that it is more effective to use a drug that can be applied directly to the nose as opposed to a pill that would take time to work
- However, when you have a cold you have other symptoms that cannot be relieved if you use a spray or inhaler that acts too locally. May prefer to take a pill (systemic) so that you can relieve other symptoms (e.g. nasally-applied drugs may not unblock Eustacian tubes).
- Some side effects can be beneficial and can be exploited
- Not all allergy sufferers can be helped by antihistamines (30% not adequately helped)