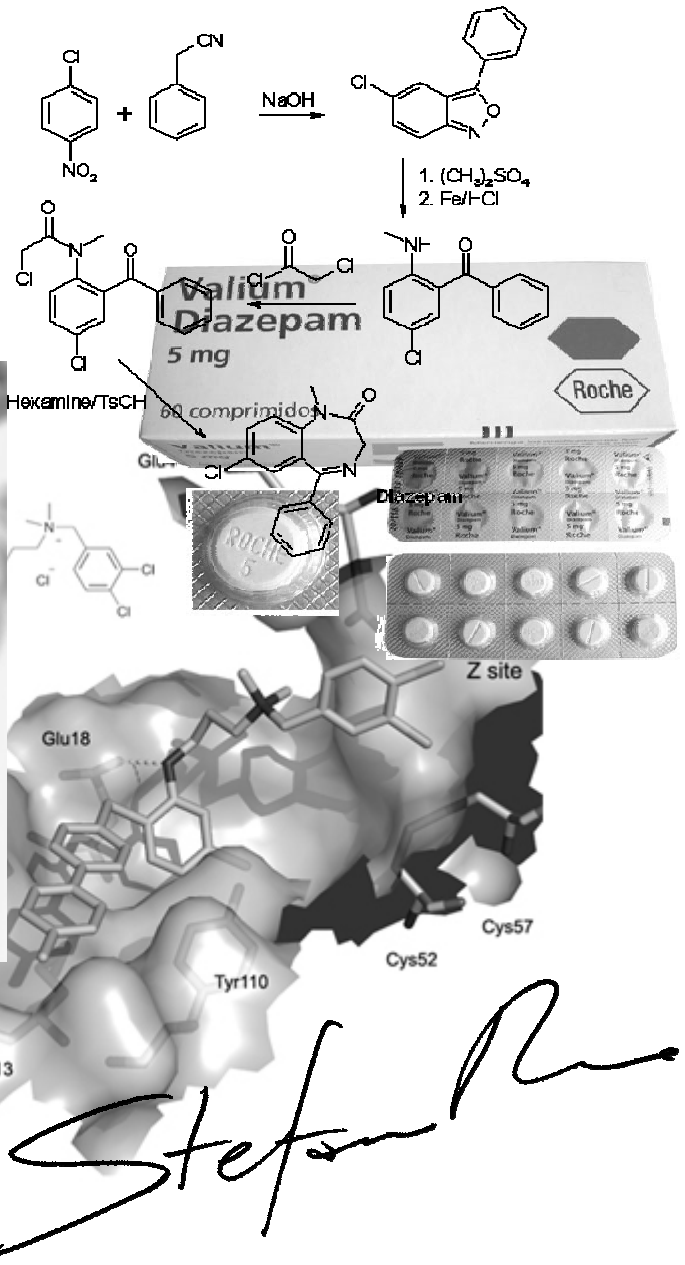
Chimica Farmaceutica e Tossicologica – Parte II



Histaminergic system and antiulcer drugs

Parte I

1. Introduction

1.1 Ulcers

- Localised erosions of the mucous membranes of the stomach and duodenum
- Potentially fatal if untreated
- Caused by stress, infection (*H. Pylori*) and drugs (NSAIDS)
- Aggrevated by gastric acid (HCl) in the stomach

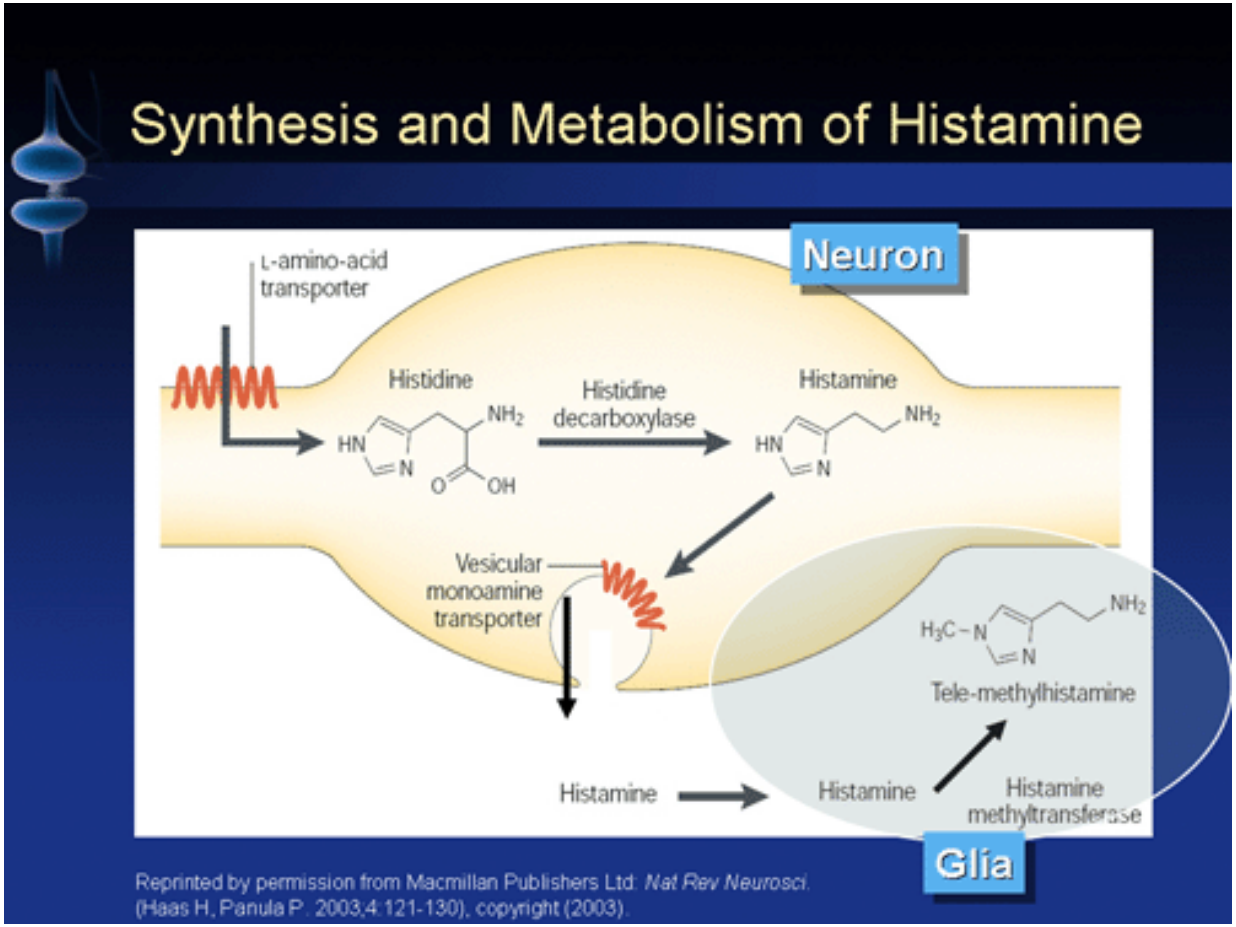
1.2 Therapy of ulcers

- Lower the levels of gastric acid

  -histamine antagonists and proton pump inhibitors
- Antibacterial agents vs. *H. Pylori*
- Herbal remedies

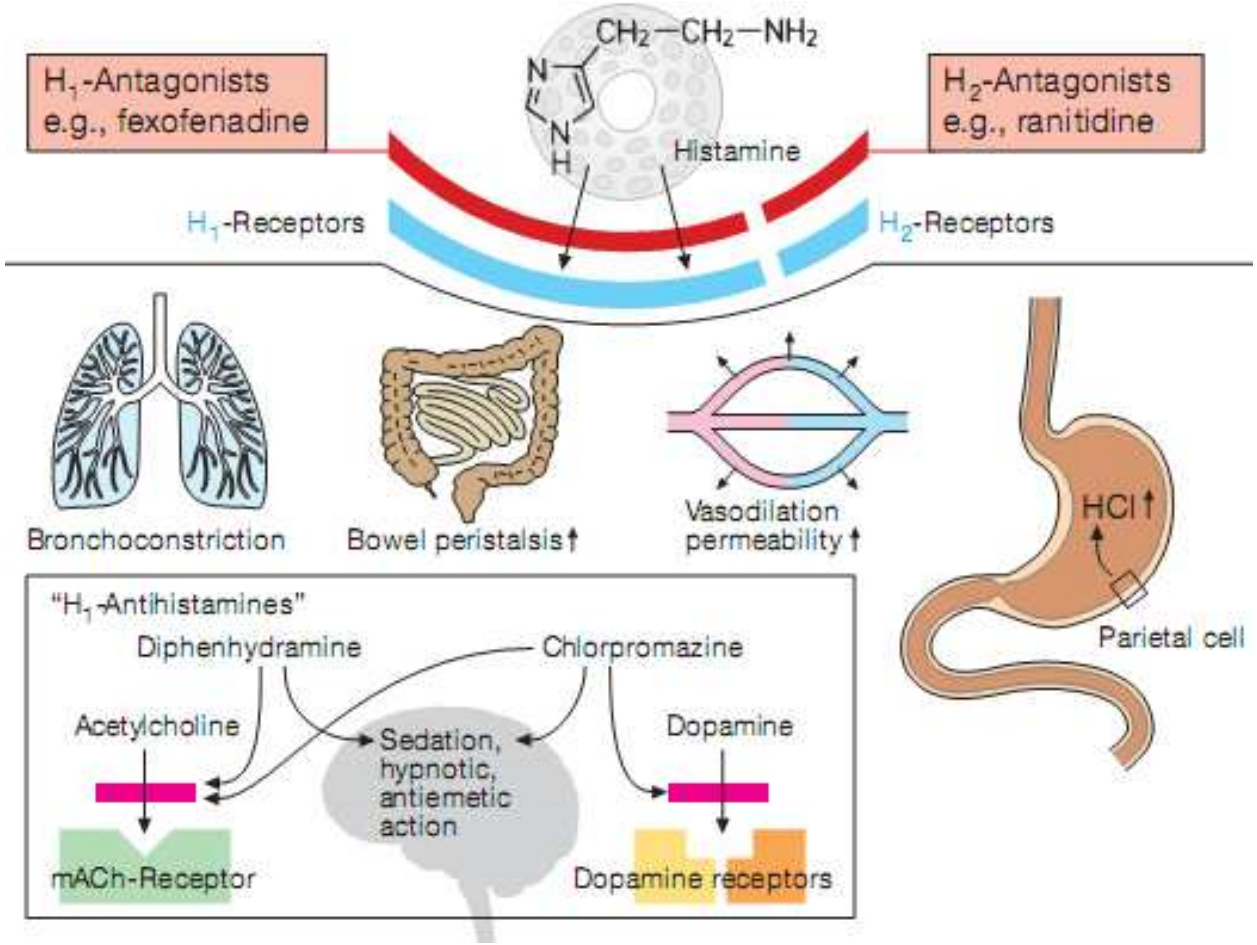
1. Introduction

1.1 Biosynthesis



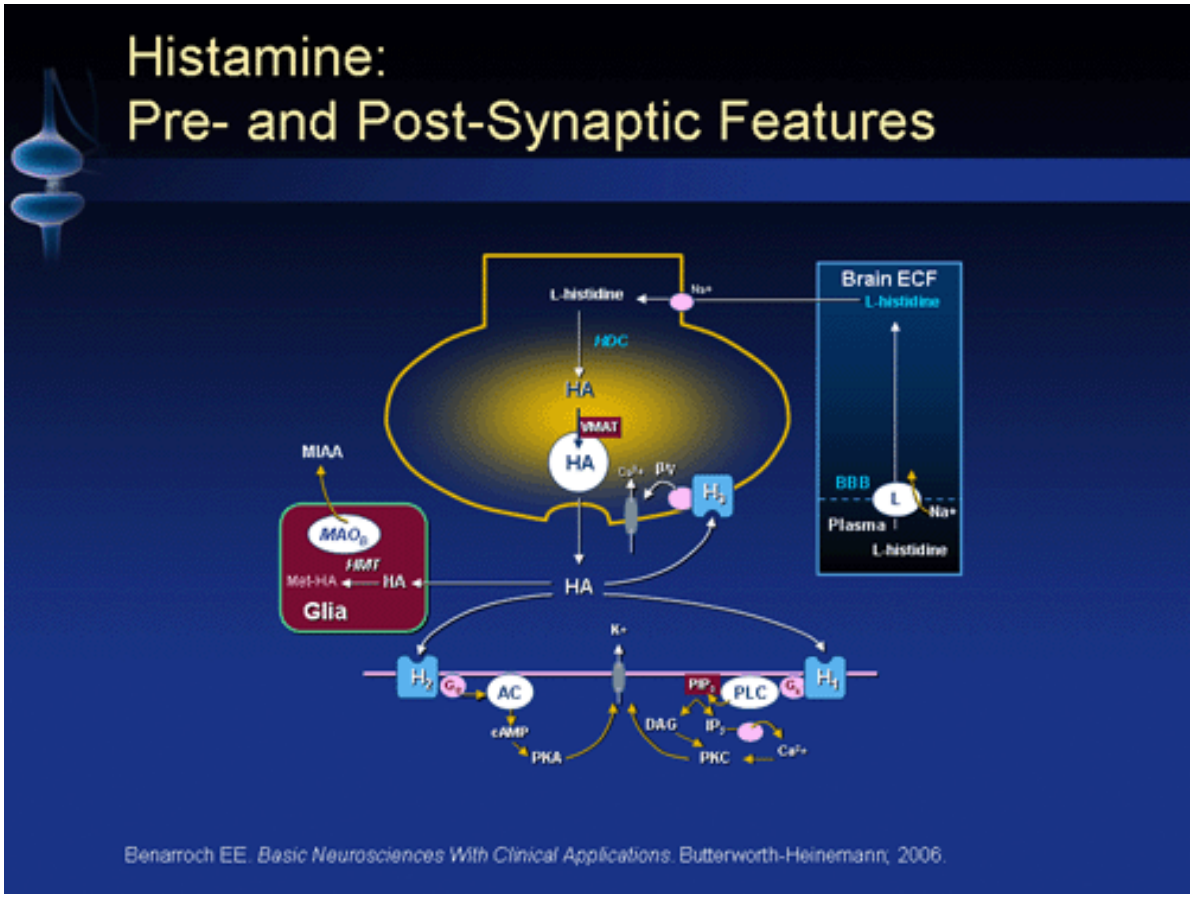
1. Introduction

1.3 Histamine effetcts



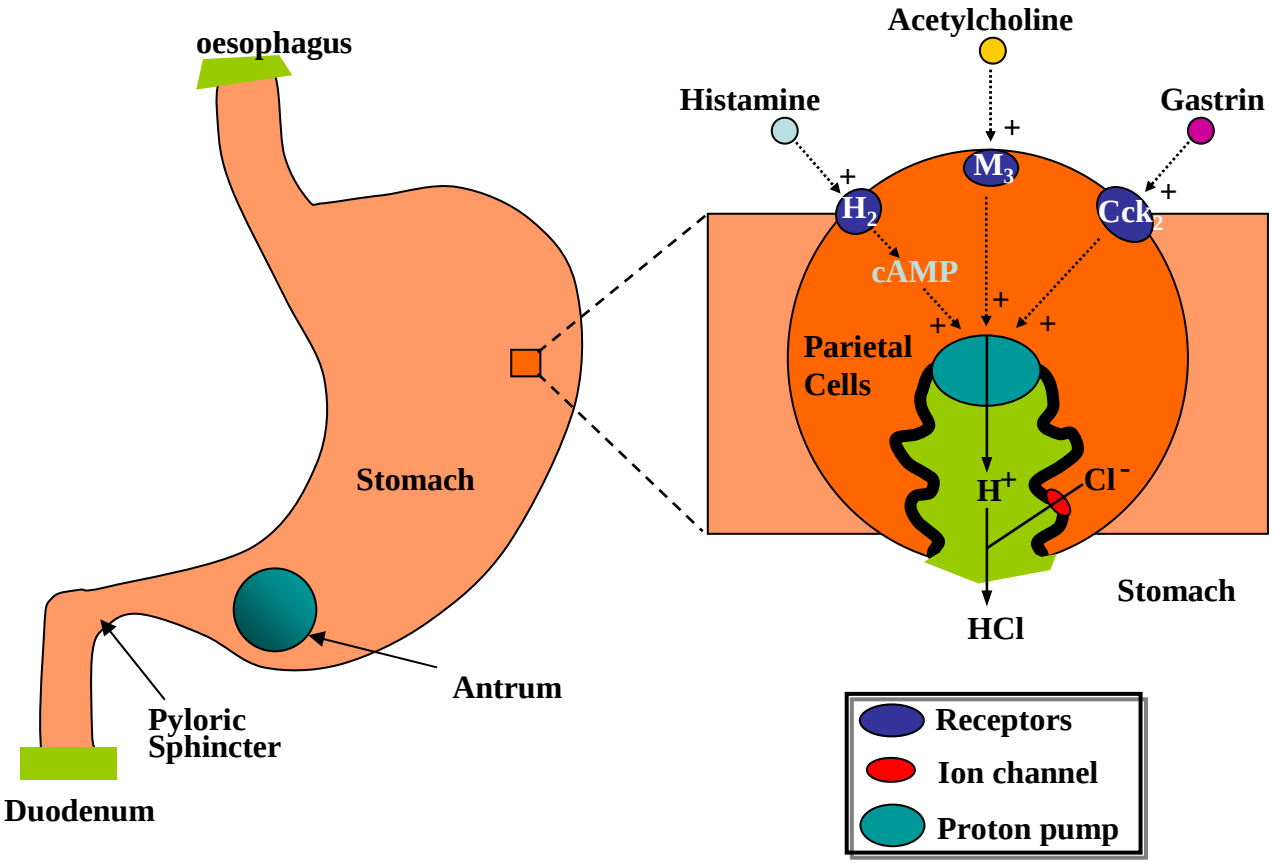
1. Introduction

1.3 Histamine effetcts



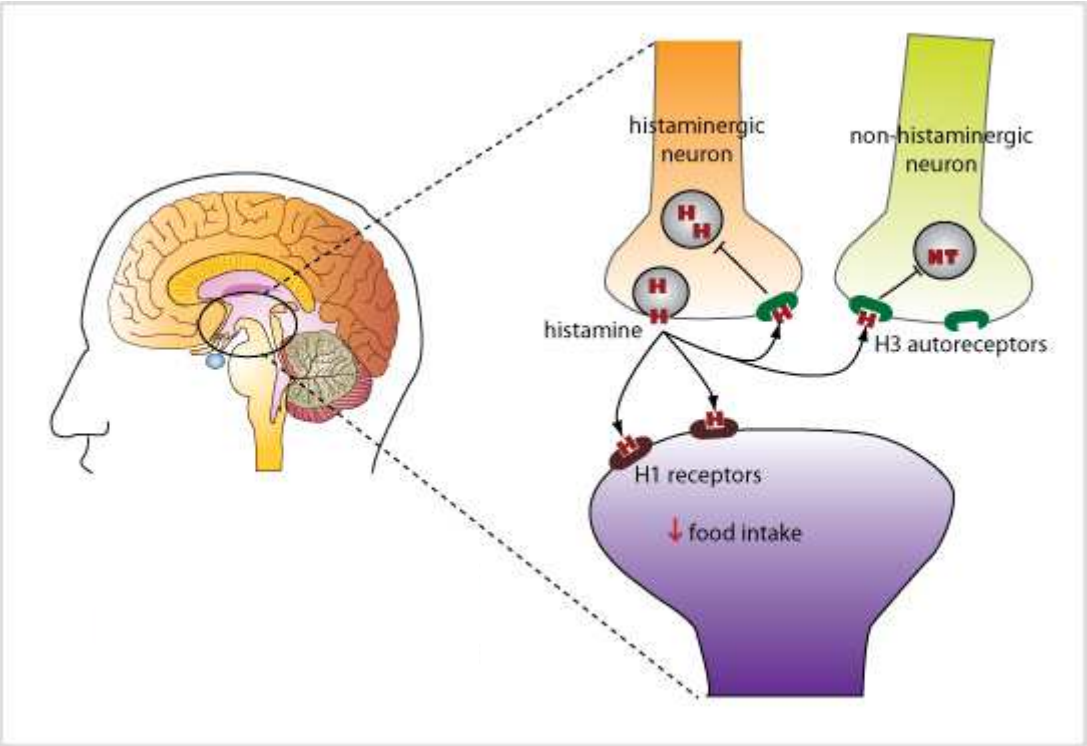
1. Introduction

1.3 Parietal cells and gastric acid release



- **Release of gastric acid is promoted by acetylcholine, gastrin and histamine**

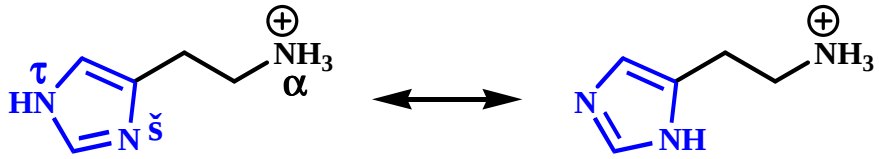
1. Introduction
1.4 SNC histaminergic control



2. Histamine

2.1 Properties

- A chemical messenger released by cells
- Acts as a local hormone



Imidazole ring

- Two possible tautomers
- pK_a for the α-NH_2 group = 9.80.
- % ionisation at pH 7.4 = 99.6
- pK_a for the imidazole ring = 5.74
- Imidazole ring is not ionised at blood pH

2. Histamine

2.2 Actions

Histamine is released by cell damage

↓

Stimulates dilation of blood vessels with increased permeability

↓

White blood cells escape blood vessels and access area of tissue damage

↓

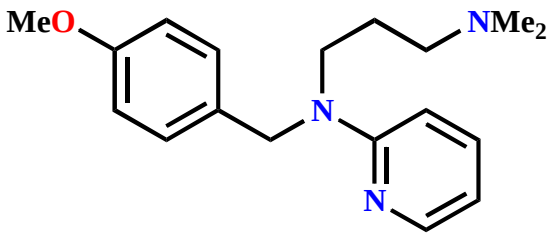
White blood cells combat infection

BUT

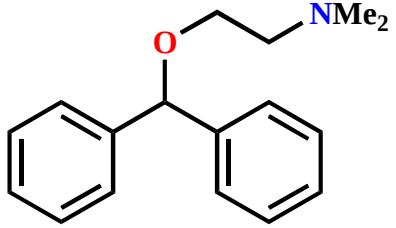
Also released by allergies, asthma, hay fever and insect bites

3. Classical Antihistamines

Commonly used to treat symptoms such as inflammation & itching



Mepyramine



Diphenhydramine
Benadryl

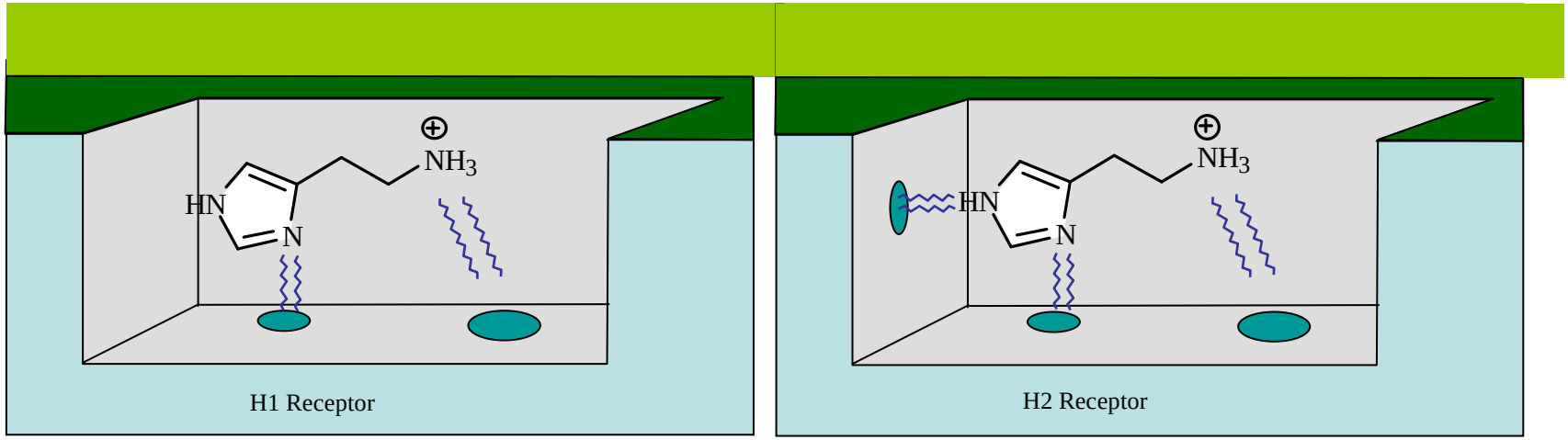
- **But no effect on gastric acid release**
- **Casts doubt on histamine receptors being present on parietal cells**
- **Histamine may promote gastric acid release indirectly**
- **SK&F propose two types of histamine receptor (H_1 and H_2)**
- **H_1 - responsible for classical actions of histamine**
- **H_2 - proposed as the receptor on the parietal cells**
- **Claim that H_2 receptors are unaffected by classical antihistamines**
- **Implies classical antihistamines are H_1 specific**

4. Histamine as a Lead Compound

- No known H_2 antagonist at the time - no lead compound
- SK&F decide to use histamine itself as the lead compound
- Aim is to alter an agonist into an antagonist
- Compare development of propranolol (β-blocker) from adrenaline
- Need to know SAR requirements for H_2 agonists
- Analogues tested by their ability to promote gastric acid release
- Does not prove existence of H_2 receptor

5. SAR for H_1 and H_2 Agonists

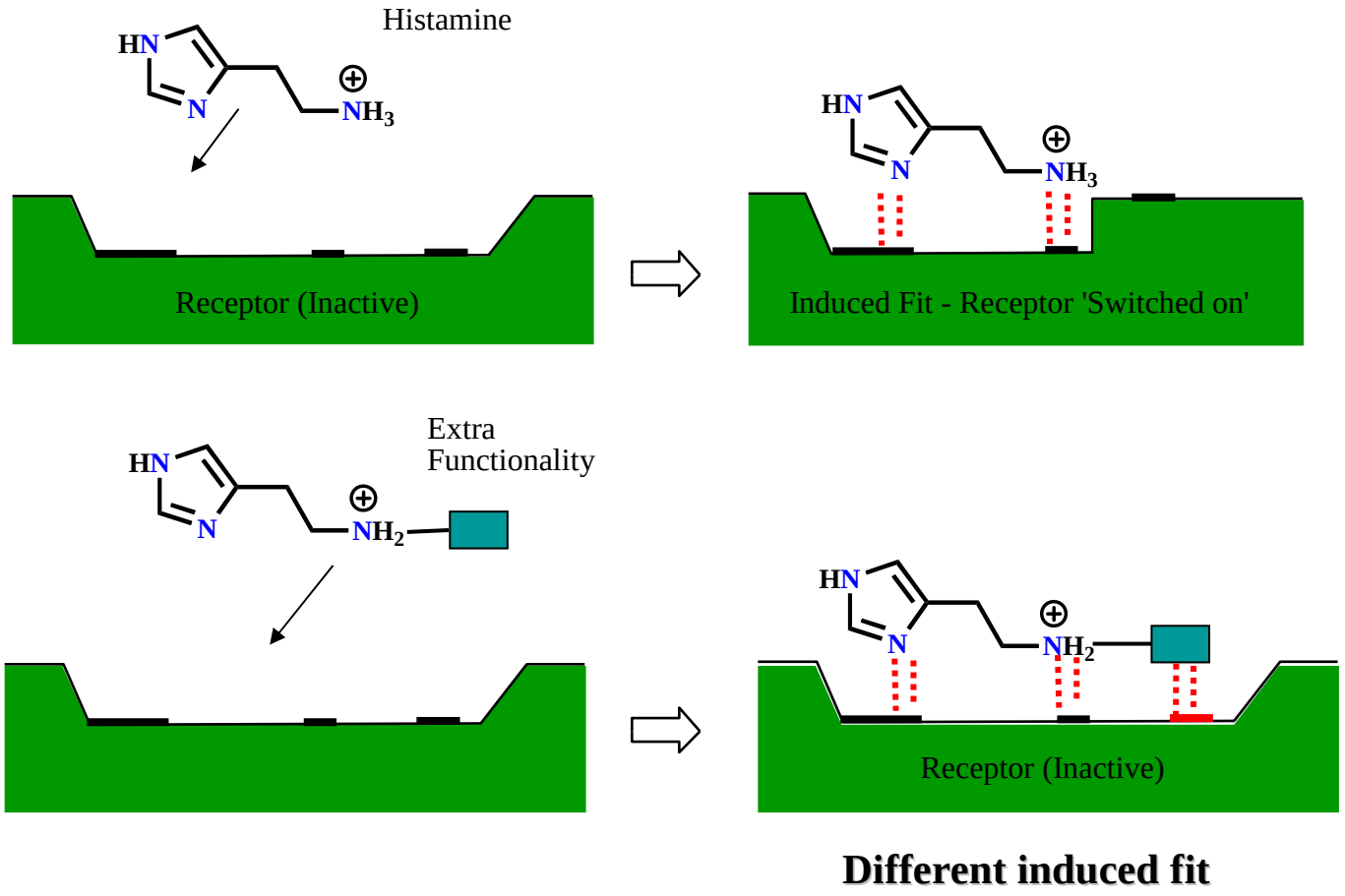
- **Two nitrogen atoms are required for H_1 agonist activity**
- **All three nitrogen atoms are required for H_2 agonist activity**



6. Strategies for converting Agonists to Antagonists

- **Add extra functional groups to find extra binding interactions with the binding site**
- **Extra binding interactions may result in a different mode of binding resulting in a different induced fit for the receptor**
- **Different induced fit may fail to activate the receptor**
- **A a result, analogue binds but fails to activate the receptor**
- **Analogue likely to bind more strongly than an agonist**

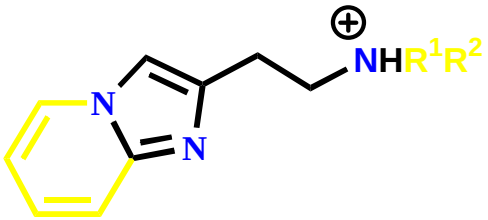
6. Strategies for converting Agonists to Antagonists

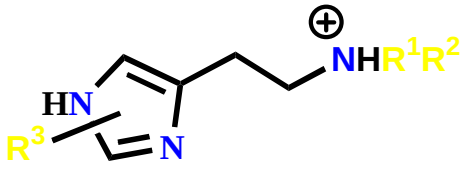


Different induced fit

6. Strategies for converting Agonists to Antagonists

Examples - extra hydrophobic groups

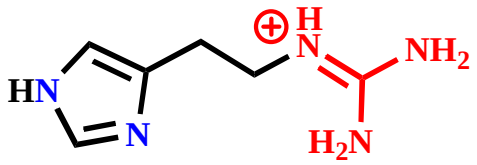




Results

- No antagonist activity observed with extra hydrophobic groups
- Try adding extra hydrophilic groups instead
- Aim is to search for extra polar binding regions

7. N^{a}-Guanylhistamine



Guanidine moiety

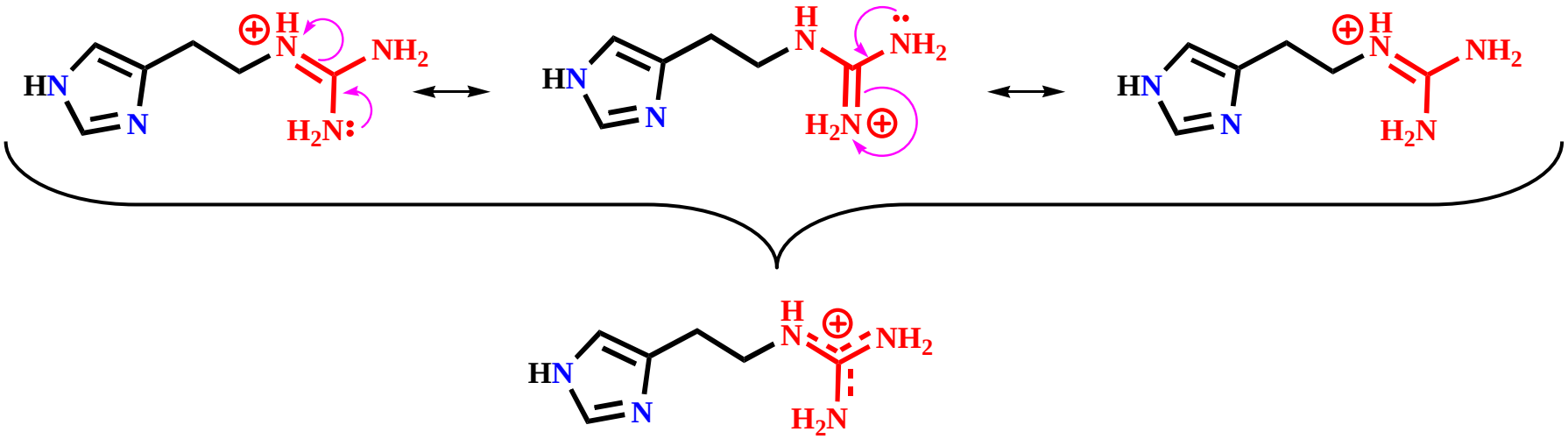
7.1 Biological properties

- **Partial agonist - promotes HCl release but less strongly than histamine**
- **Prevents histamine from fully promoting the release of HCl**
- **SK&F suggest that N^{α}–guanylhistamine is binding to the proposed H_2 receptor, resulting in weak activation**
- **Whilst present, N^{α}–guanylhistamine blocks histamine from binding**

7. N^a-Guanylhistamine

7.2 Structure and chemical properties

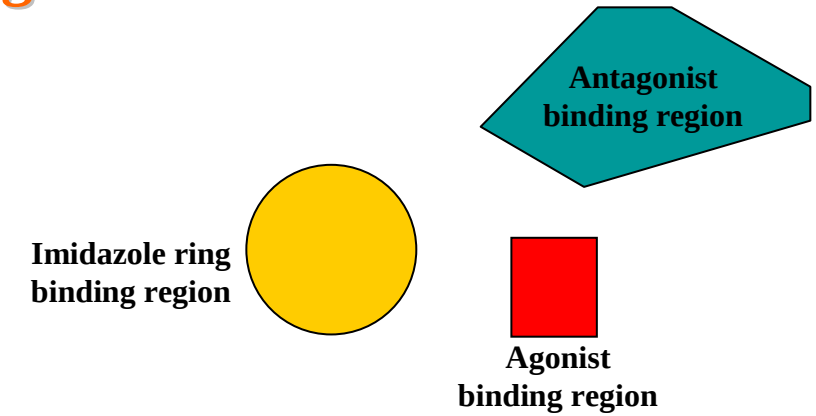
- **The guanidine group is basic and ionised**
- **Different tautomers are possible**
- **The positive charge can be delocalised**



The positive charge is more diffuse and can be further away from the imidazole ring

8. Binding Theory for Agonists and Antagonists

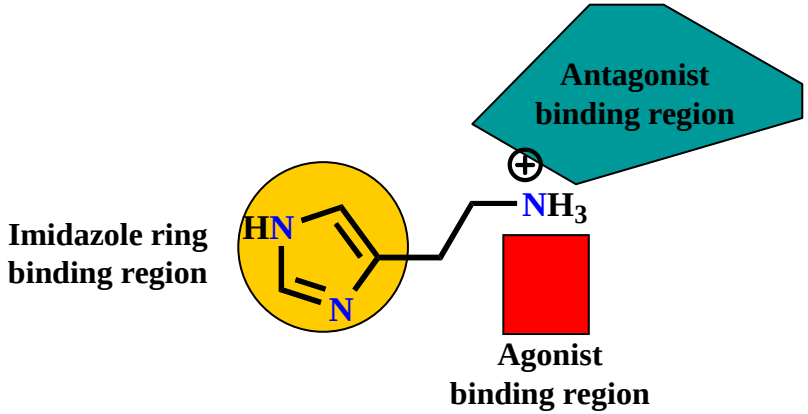
8.1 Binding regions



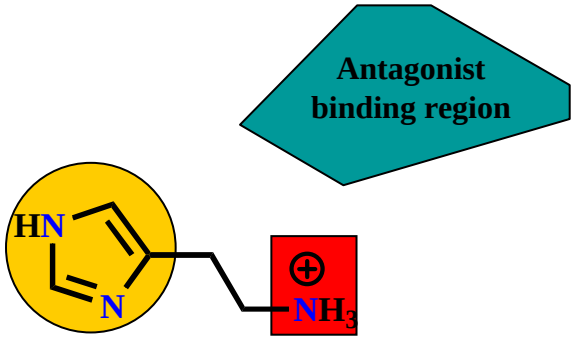
- Three binding regions are proposed for the H_2 receptor - an imidazole binding region and two polar binding regions
- Two binding modes are proposed - one for agonists and one for antagonists
- The imidazole binding region is common to both binding modes
- One of the polar binding regions is accessed by agonists and the other by antagonists
- The antagonist polar region is further from the imidazole binding region

8. Binding Theory for Agonists and Antagonists

8.2 Binding of histamine



No interaction as an antagonist

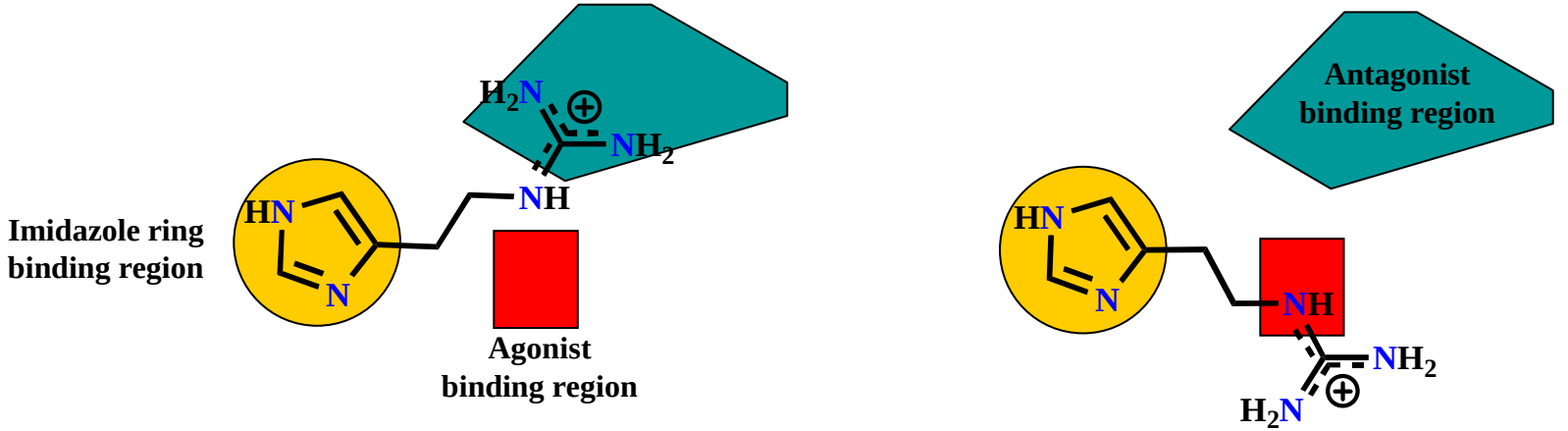


Strong interaction as an agonist

- Histamine has a short chain
- Charged α-nitrogen can only reach the polar agonist region
- The antagonist binding region is out of range
- Histamine can only bind as an agonist
- Histamine acts as a pure agonist

8. Binding Theory for Agonists and Antagonists

8.3 Binding of *N*α-guanylhistamine



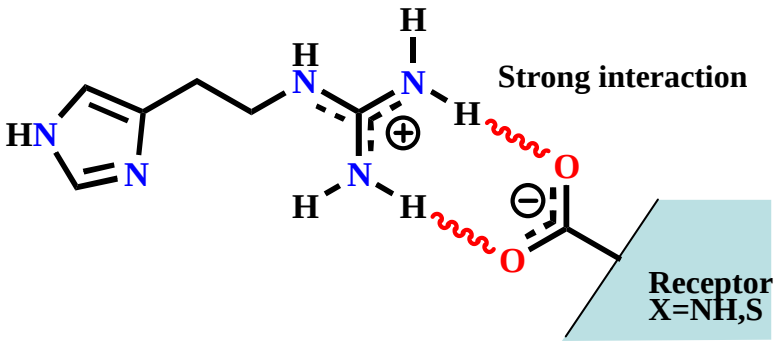
Binding as an antagonist
Receptor not activated

Binding as an agonist
Receptor activated

- **Positive charge on the structure is more diffuse and further out**
- **Allows *N*α-guanylhistamine to bind in two different modes**
- **Structure binds as an agonist in one mode and as an antagonist in the other mode, making it a partial agonist**

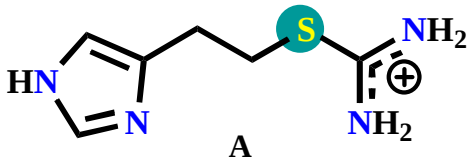
9. Chelation Binding Theory

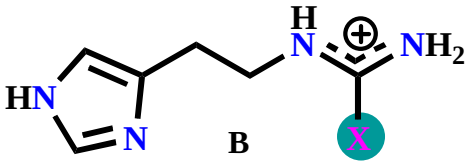
9.1 The proposal



SK&F propose that the guanidine moiety interacts with a carboxylate ion in the antagonist binding region by means of two H-bonds and an ionic interaction

9.2 The evidence



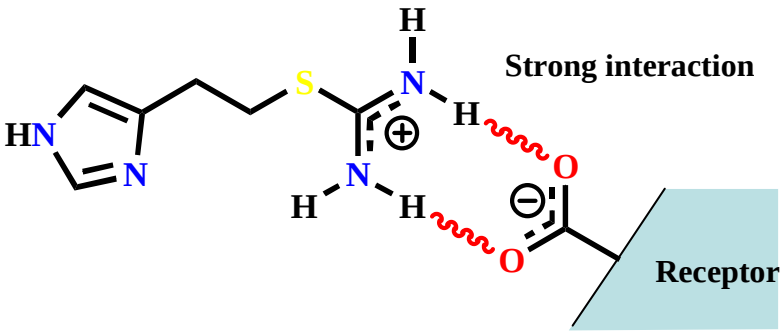


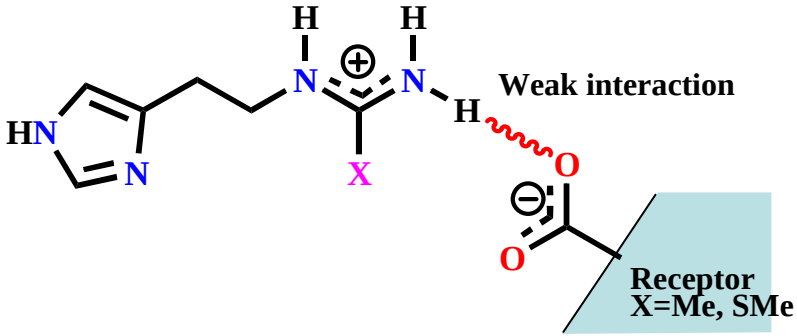
X= SMe, Me

Structures A and B are both partial agonists, but structure A has greater antagonist properties

9. Chelation Binding Theory

9.3 Binding modes for analogues





Positive charge is localised further out leading to better interactions with the antagonist binding region

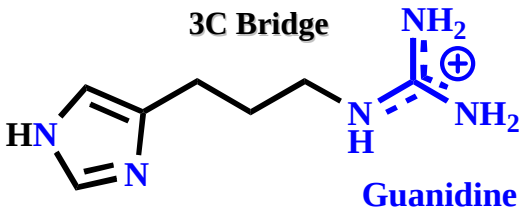
Only one H-bond is possible with the antagonist binding region. Charge is also directed away from the carboxylate ion - weaker antagonist property.

- **The chelation binding theory was eventually disproved but it served a purpose in explaining results and pushing the project forward on rational grounds**

10. Chain Extension Strategy

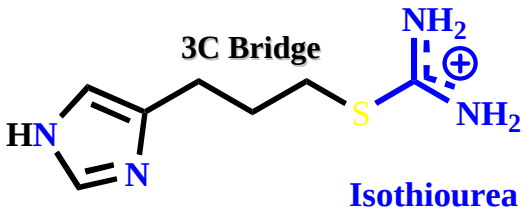
10.1 Aim: To push the polar guanidine group further out and to increase the interaction with the antagonist binding region

10.2 Results:



Guanidine

Partial agonist
Antagonist activity increases



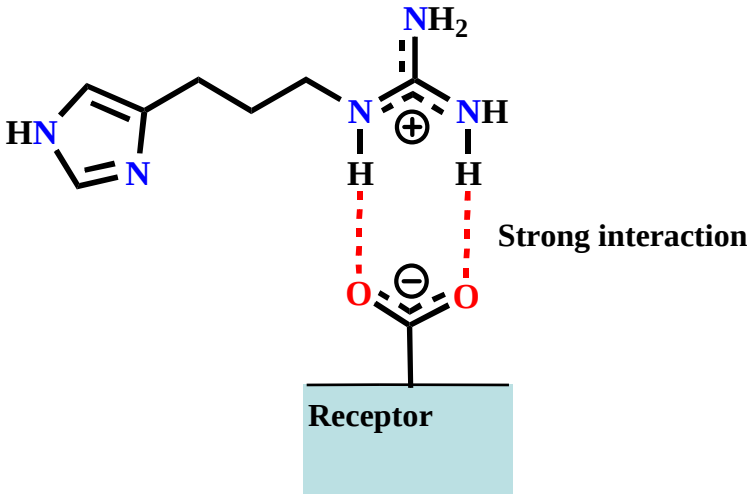
Isothiourea

Partial agonist
Antagonist activity decreases!

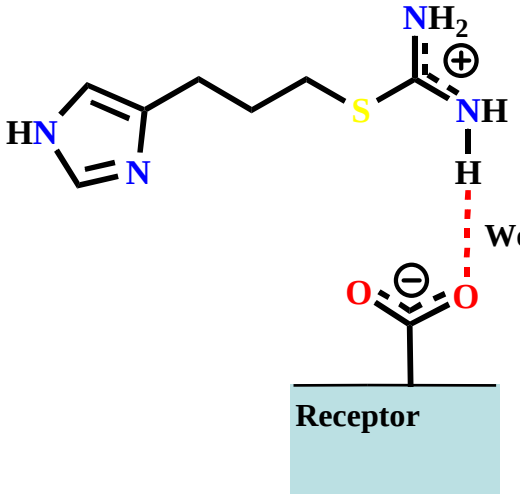
- **Antagonist activity of the extended guanidine analogue increases as expected**
- **Isothiourea analogue might have been expected to have increased antagonist activity since the charge is further out**

10. Chain Extension Strategy

10.3 Proposed binding for 3C extension analogues



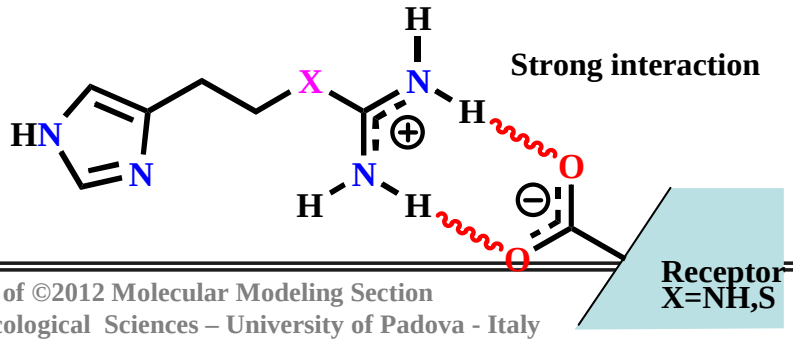
Strong interaction



Weak interaction

- **Different form of hydrogen bonding taking place**

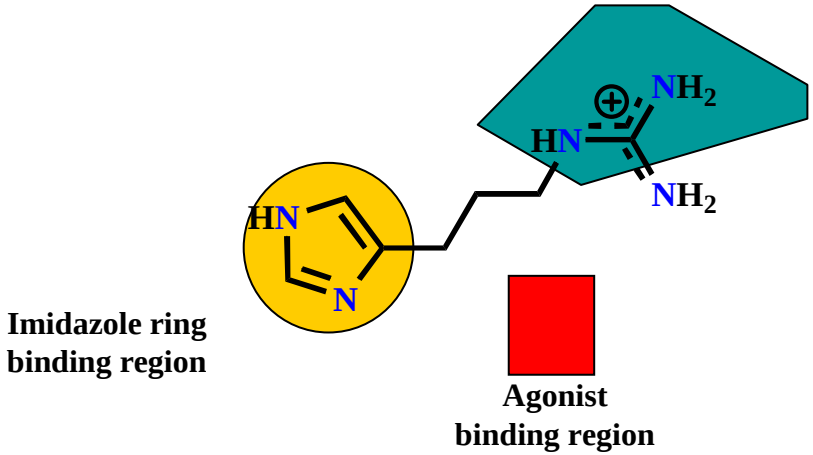
Compare 2C bridged analogues



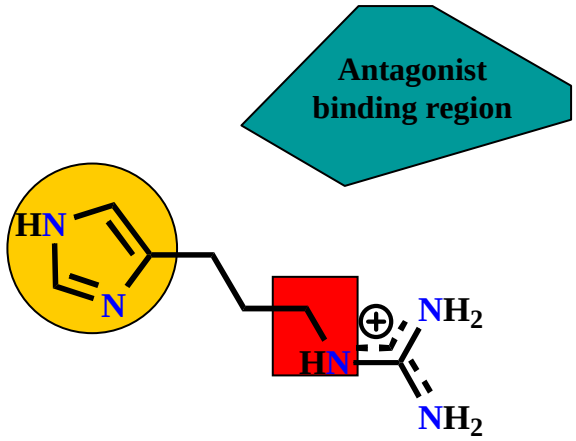
Strong interaction

10. Chain Extension Strategy

10.3 Proposed binding for 3C extension analogues



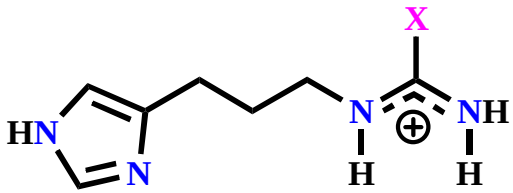
Good binding as an antagonist



Binding as an agonist

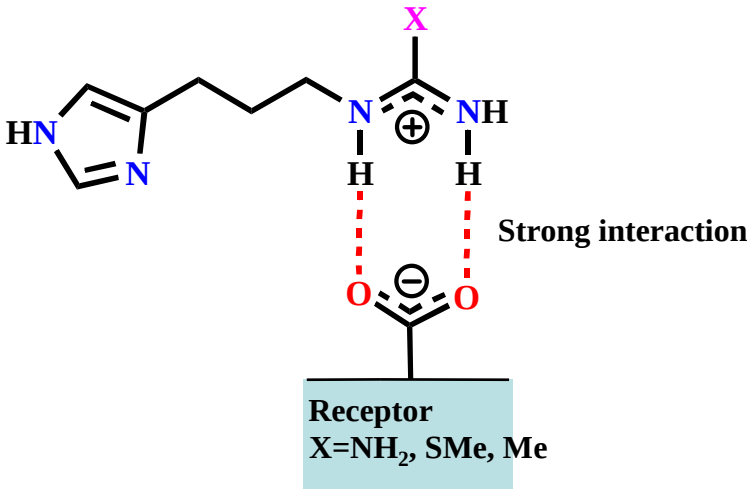
10. Chain Extension Strategy

10.4 Further evidence



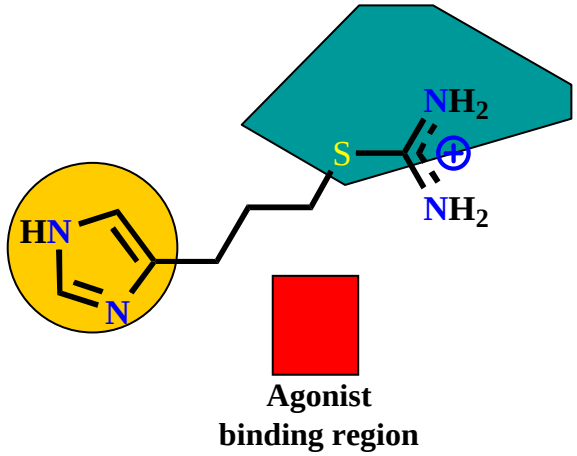
Partial agonists with good antagonist activity (X= Me or SMe)

Binding mode

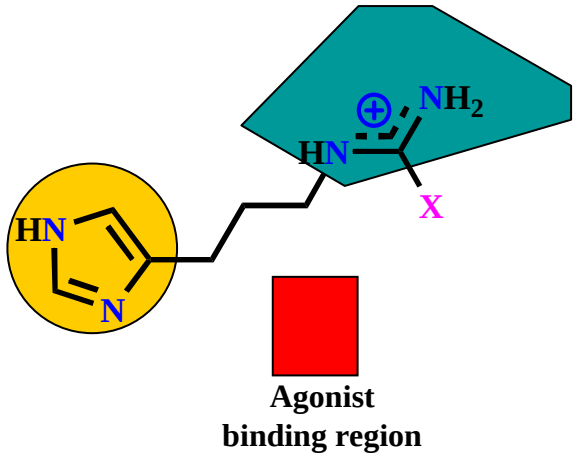


10. Chain Extension Strategy

10.4 Further evidence



Poor binding as an antagonist



Good binding as an antagonist

- **Emphasis now switches to the types of binding interactions at the polar binding regions**

11. Distinguishing between the Polar Binding Regions

11.1 Strategy:

- **Replace the ionic guanidine group with a neutral H-bonding group**

11.2 Rationale:

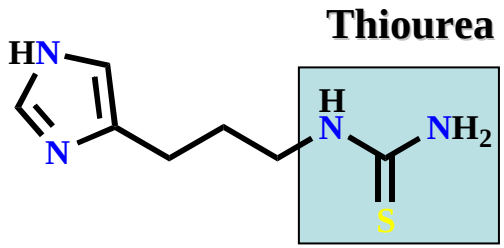
- **May allow a distinction to be made between the two polar binding regions.**
- **Ionic bonding is known to be crucial for the agonist binding region**
- **It may not be crucial for the antagonist binding region**

11.3 Method:

- **Replace the basic guanidine moiety with a neutral thiourea group**

11. Distinguishing between the Polar Binding Regions

11.4 SK&F 91581



No agonist activity, very weak antagonist

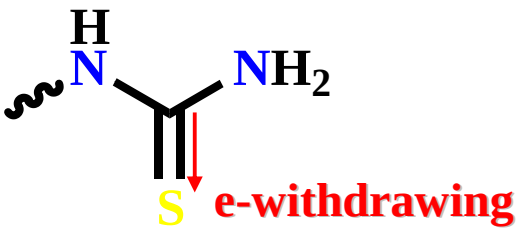
11. Distinguishing between the Polar Binding Regions

11.5 Comparison between the thiourea and guanidine groups

Similarities - Planarity, geometry, size, polarity, H-bonding ability

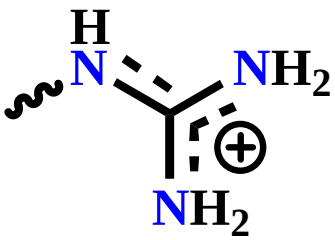
Differences - Thiourea is neutral while guanidine is basic and ionised

Thiourea



Neutral

Guanidine



Basic

Conclusions -

- **Agonist polar region involves ionic and H-bonding interactions**
- **Antagonist polar region may not require ionic interactions. H-bonding may be sufficient**

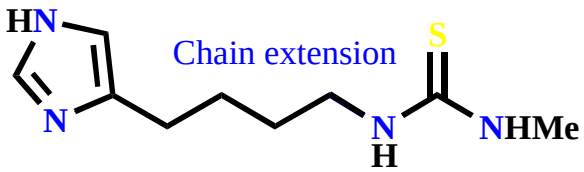
12. Chain Extension

Strategy

- **Extend the carbon bridge to 4 carbons**
- **Pushes thiourea group further out**
- **May increase the interaction with the antagonist binding region**

Results

Discovery of burimamide



12. Chain Extension

Properties of burimamide

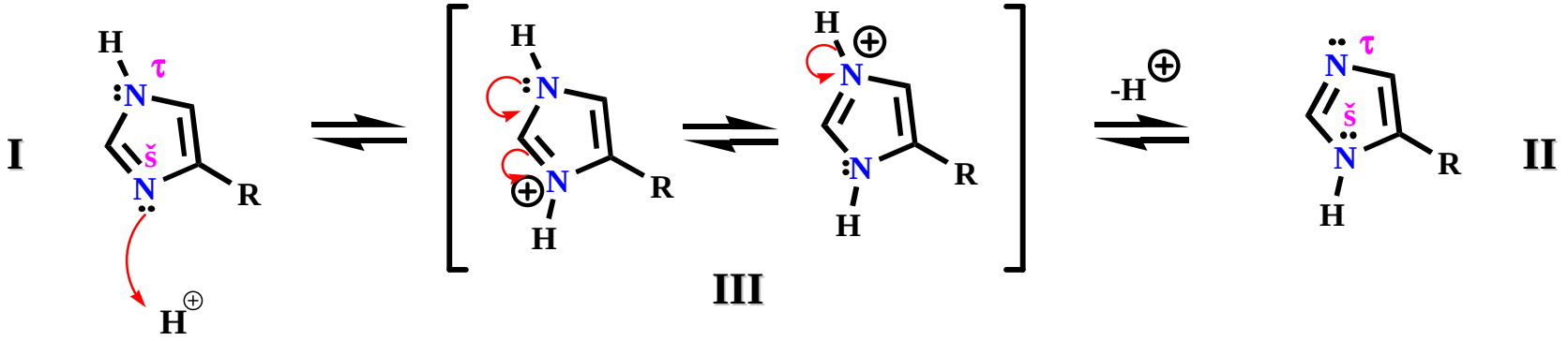
- 100 times more active as an antagonist compared to N^{α}-guanylhistamine
- No antagonist activity at H_1 receptors
- Activity too low for oral use

Conclusions –

- Chain extension leads to a pure antagonist with good activity
- Chain extension allows a better overlap of the thiourea group with the antagonist binding region
- Establishes the existence of H_2 receptors

13. The Imidazole Ring

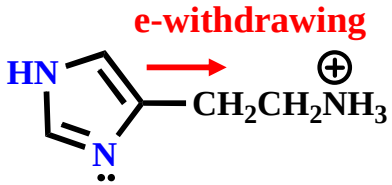
13.1 Structures



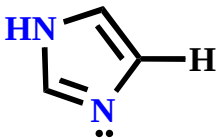
- **Imidazole ring can exist as two tautomers (I) and (II) as well as two ionised forms (III)**
- **Which of these is preferred?**

13. The Imidazole Ring

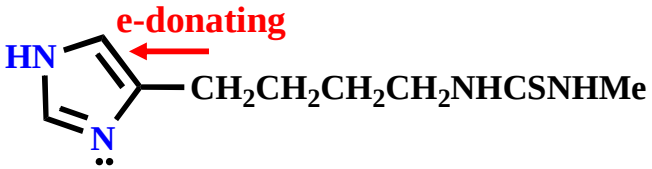
13.2 Basicity



Histamine
pK_a = 5.74
Ionisation = 3%



Imidazole
pK_a = 6.80



Burimamide
pK_a = 7.25
Ionisation = 40%

Conclusions

- **The imidazole ring of histamine is not ionised when it interacts with the imidazole binding region**
- **The ionised form of burimamide is unlikely to bind well**
- **Decreasing the basicity and ionisation of the imidazole ring in burimamide closer to that of histamine may increase the binding interactions to the imidazole binding region**

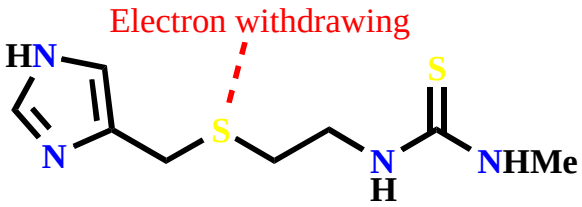
13. The Imidazole Ring

13.3 Varying basicity

Strategy

Convert the side chain of burimamide to an e-withdrawing group

Thiaburimamide

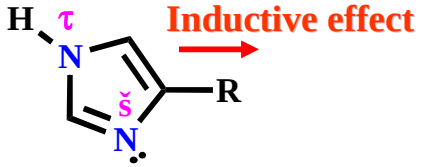


pK_a = 6.25
Increase in antagonist activity
Non-ionised imidazole is favoured

13. The Imidazole Ring

13.4 Tautomer studies

Tautomer I vs tautomer II



- Favoured tautomer for histamine is I
- Side chain is electron withdrawing
- Inductive effect decreases with distance
- N^{π} is less basic than N^{τ}
- N^{τ} is more likely to be protonated
- Favoured tautomer for thiaburimamide is also tautomer I

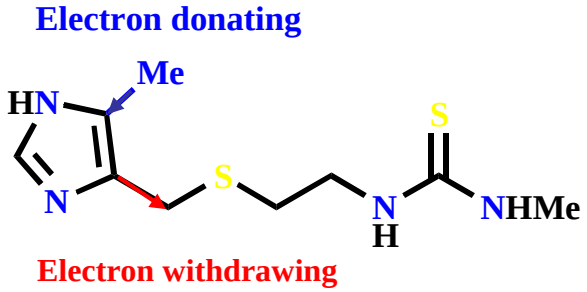
Strategy

- Increase the basicity of N^{τ} relative to N^{π} to further increase the percentage population of tautomer I vs tautomer II
- Add an electron donating group to the imidazole ring closer to N^{τ} than to N^{π}

13. The Imidazole Ring

13.4 Tautomer studies

Metiamide



- 10 fold increase in antagonist activity w.r.t burimamide
- Electron-donating effect of methyl group is more significant at N^{τ}
- Increases basicity of N^{τ}
- Favours tautomer I over tautomer II
- Increase in pK_a to 6.80
- Increase in ionisation to 20%
- Increase in the population of tautomer (I) outweighs the increase in population of the ionised structures (III)
- Unacceptable side effects - kidney damage

14. Alternative Rationales

- **The increases in activity for thiaburimamide and metiamide may be due to a conformational effect**

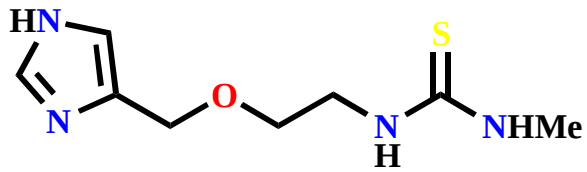
- **The thioether link increases the length and flexibility of the side chain**

- **This may lead to increased binding**

- **The methyl substituent may orientate the side chain into the active conformation - i.e. the methyl group acts as a conformational blocker**

14. Alternative Rationales

Oxaburimamide



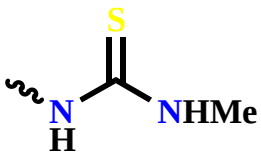
- **Less potent than burimamide despite the side chain being electron withdrawing**

Possible explanations

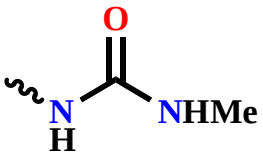
- **The ether link is smaller and less flexible**
- **The ether may be involved in a 'bad' hydrogen bond**
- **There may be an energy penalty involved in desolvating the oxygen prior to binding**

15. From Metiamide to Cimetidine

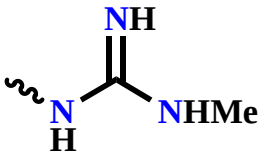
- The side effects of metiamide may be due to the thiourea group
- The thiourea group is not a natural functional group
- Replacing thiourea with a natural functional group may remove the side effects



Thiourea
Toxic side effects



Urea
Drop in activity



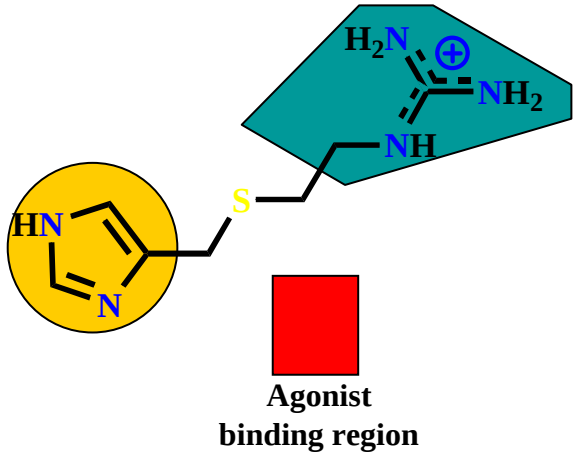
Guanidine
Drop in activity but no agonist activity!

Conclusions

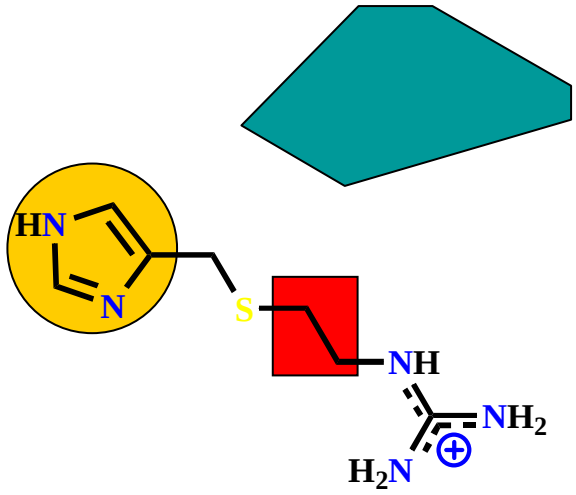
- First guanidine analogue to be a pure antagonist
- The longer 4C chain pushes the guanidine unit beyond the agonist binding region, but not beyond the antagonist binding region

15. From Metiamide to Cimetidine

Binding interactions for the 4C extended guanidine



Binding as an antagonist



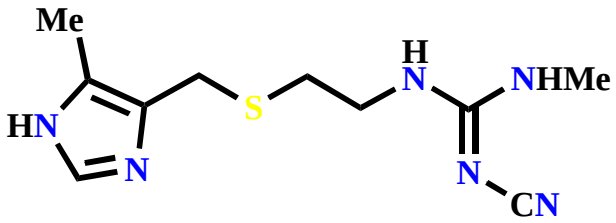
No binding as an agonist

15. From Metiamide to Cimetidine

Strategy:

- **Retain the guanidine group**
- **Guanidine is a natural group present in the amino acid arginine**
- **Increase activity by making the guanidine group neutral**
- **Add a strong electron withdrawing group to decrease basicity (e.g. NO_2 or CN)**

Cimetidine



Electron withdrawing cyanide group

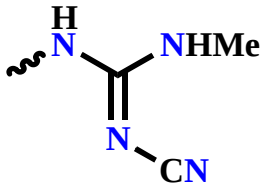
16. Cimetidine (Tagamet)

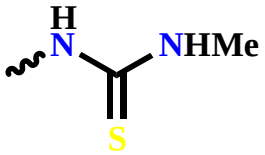
16.1 Properties

- Comparable activity to metiamide
- Less side effects
- Inhibits H_2-receptors and lowers levels of gastric acid released
- Marketed in 1976
- Biggest selling prescription drug until ranitidine
- Metabolically stable
- Inhibits cytochrome p450 enzymes
- Drug-drug interactions with diazepam, lidocaine and warfarin

16. Cimetidine (Tagamet)

16.2 The cyanoguanidine moiety



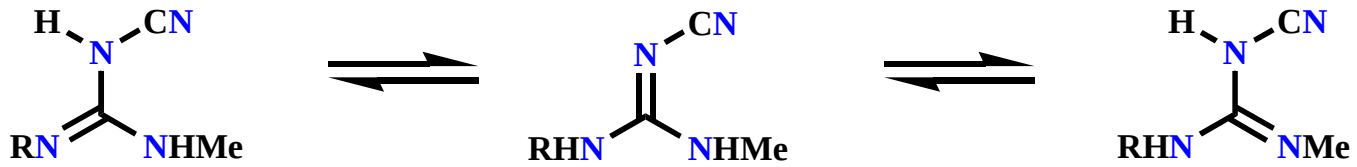


- **Acts as a bio-isostere for the thiourea group**
- **Both groups are planar and of similar geometry**
- **Both groups are polar but essentially neutral**
- **Both groups have high dipole moments**
- **Both groups have low partition coefficients**
- **The cyanoguanidine group is weakly acidic and weakly basic - amphoteric**
- **The cyanogaunidine group is not ionised at pH 7.4**

16. Cimetidine (Tagamet)

16.3 The cyanoguanidine moiety - tautomers

- **The favoured tautomer is the imino tautomer**



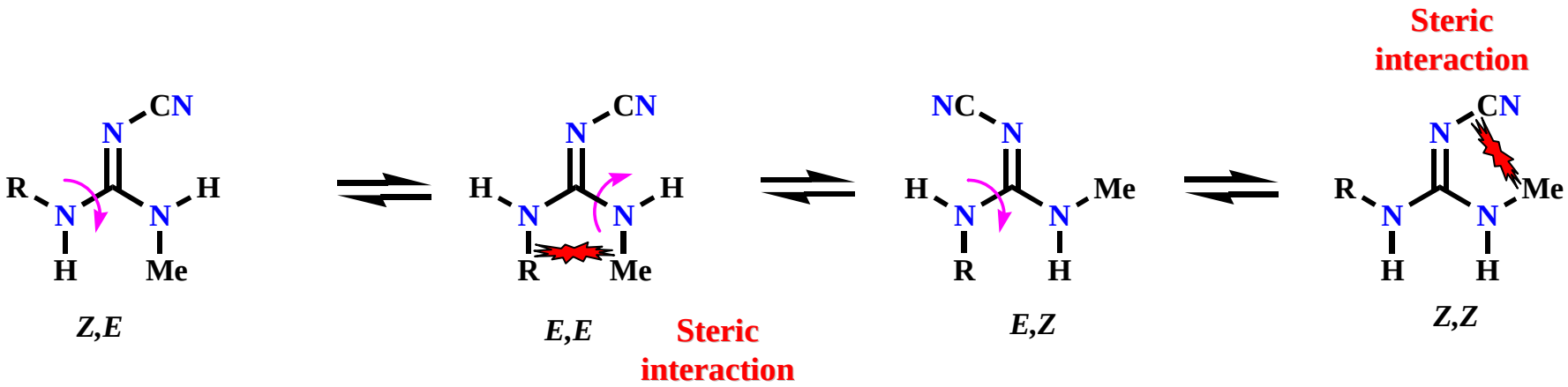
I **II** **III**

Amino Tautomer **Imino Tautomer** **Amino Tautomer**

- **The electron withdrawing effect of the CN group is an inductive effect**
- **The inductive effect is felt most at the neighbouring nitrogen**
- **The neighbouring nitrogen is least likely to form a bond to hydrogen**

16. Cimetidine (Tagamet)

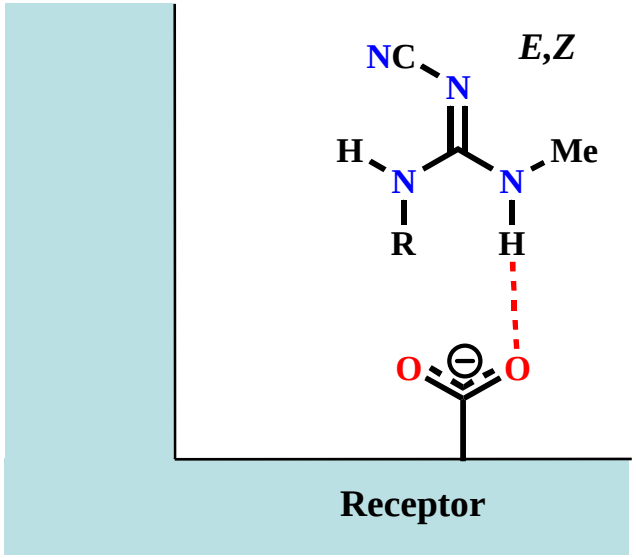
16.4 The cyanoguanidine moiety - conformational isomers



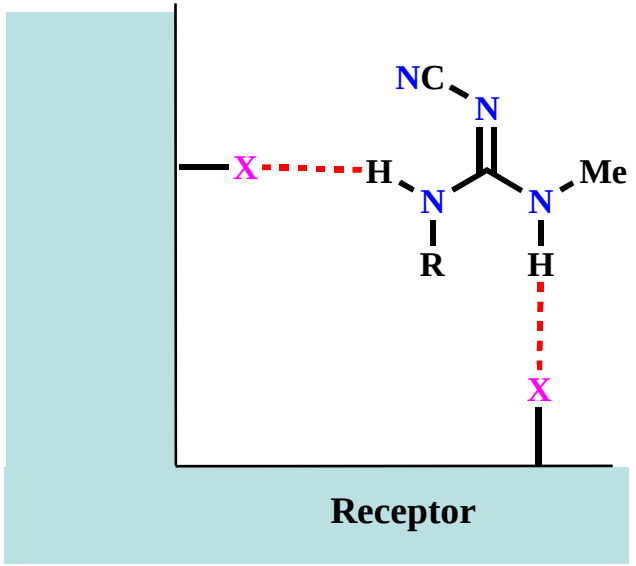
- The *E*, *E* and *Z*,*Z* conformations are not favoured - X-ray and nmr evidence
- Bad news for the chelation bonding theory
- Chelation to the one carboxylate group requires the *E*,*E* or the *Z*,*Z* conformation

16. Cimetidine (Tagamet)

16.5 The cyanoguanidine moiety - binding mode



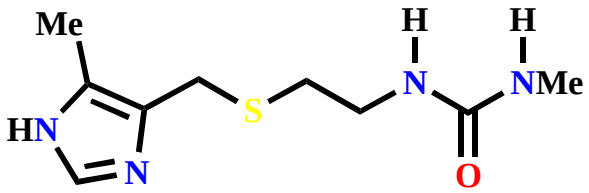
Two H-bonds are not possible for the favoured conformations



Two separate H-bonds to 2 different H-bond acceptors are more likely

17. Analogues

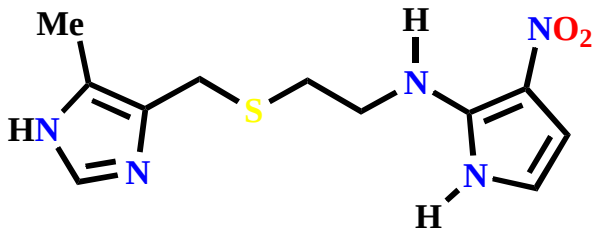
17.1 The urea analogue



- **The preferred conformation for the urea analogue is *E*,*E* or *Z*,*Z***
- **Weak antagonist**
- **Unable to bind to two different binding groups in the antagonist binding region**

17. Analogues

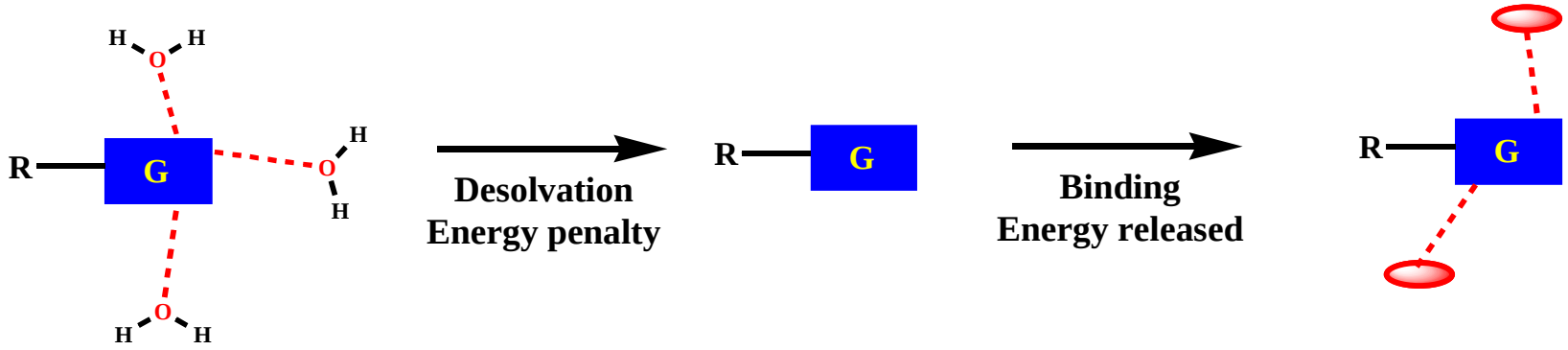
17.2 Rigid nitropyrrole analogue



- **Unable to adopt the *E*,*E* or *Z*,*Z* conformation**
- **Strongest analogue of cimetidine**
- **Locked into the active conformation**
- **Can only interact with two separate H-bond acceptors in the antagonist binding region**

18. Desolvation Theory

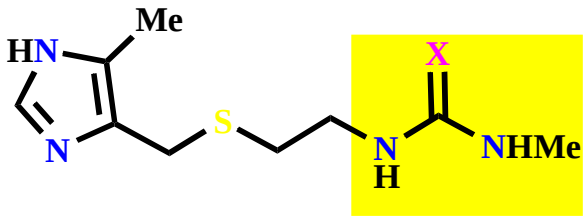
18.1 The process



- A guanidine unit is highly polar and highly solvated
- Solvated water must be removed prior to binding
- An energy penalty is involved
- The ease of desolvation may affect strength of binding and activity
- A urea group is more hydrophilic than a cyanoguanidine group
- May explain lower activity of the urea analogue

18. Desolvation Theory

18.2 Hydrophobic analogues



Aminal system (Z)

Strategy

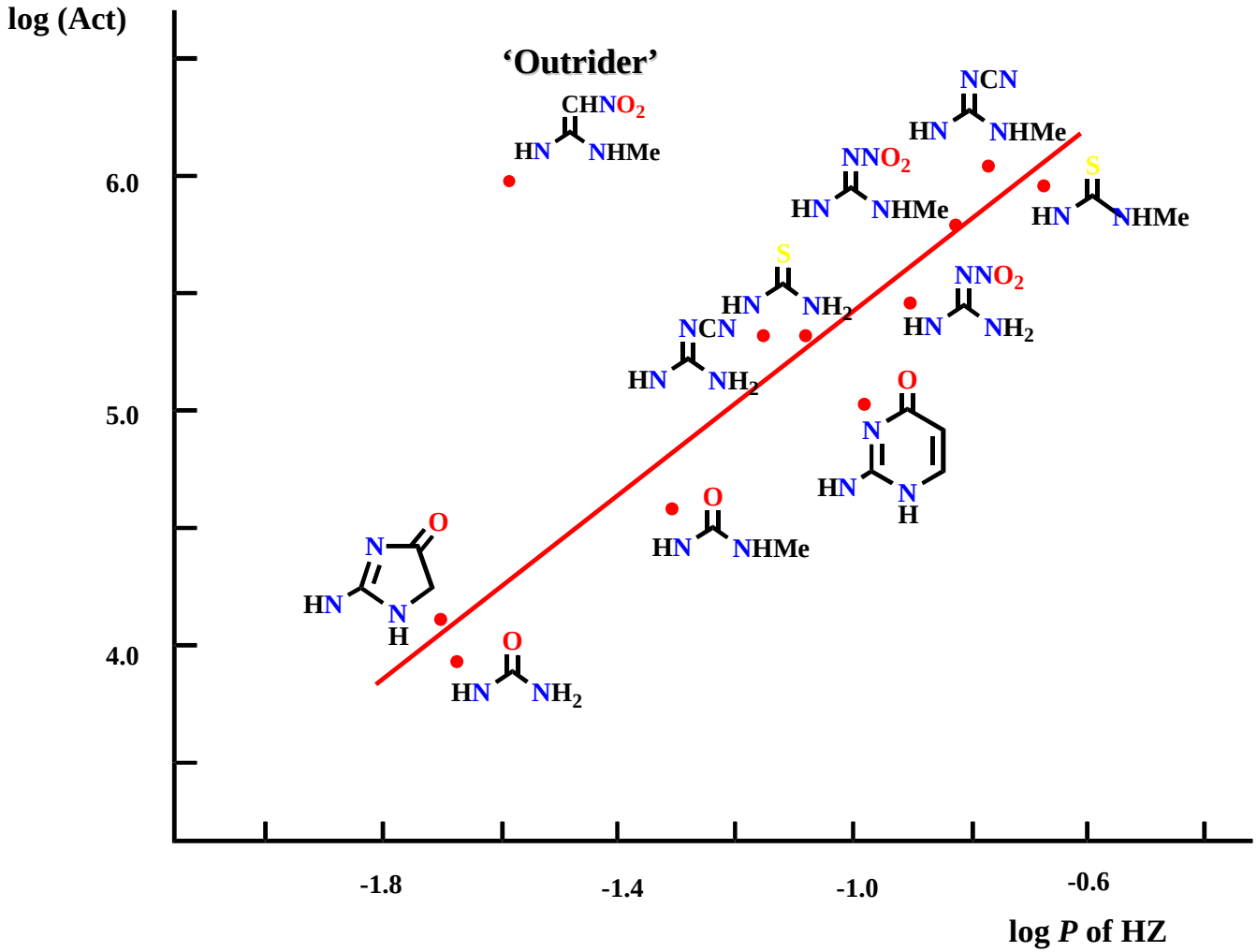
- **Increase the hydrophobic character of the planar aminal system**
- **Implies less solvation**
- **Implies less of an energy penalty associated with desolvation**
- **Implies easier binding and a stronger activity**

Result

- **Antagonist activity of analogues increases as hydrophobic character increases**

18. Desolvation Theory

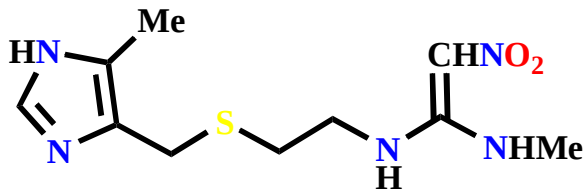
18.2 Hydrophobic analogues



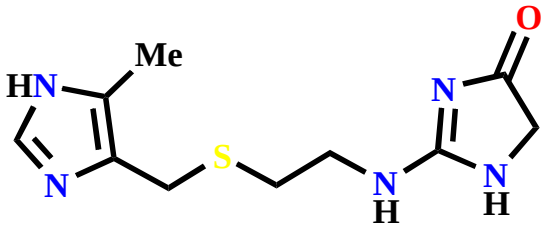
Log (activity) = 2.0 log *P* + 7.4

18. Desolvation Theory

18.2 Hydrophobic analogues



Greater activity than expected Hydrophilic group should lower activity



Lower activity than expected based on the hydrophobicity of the group present

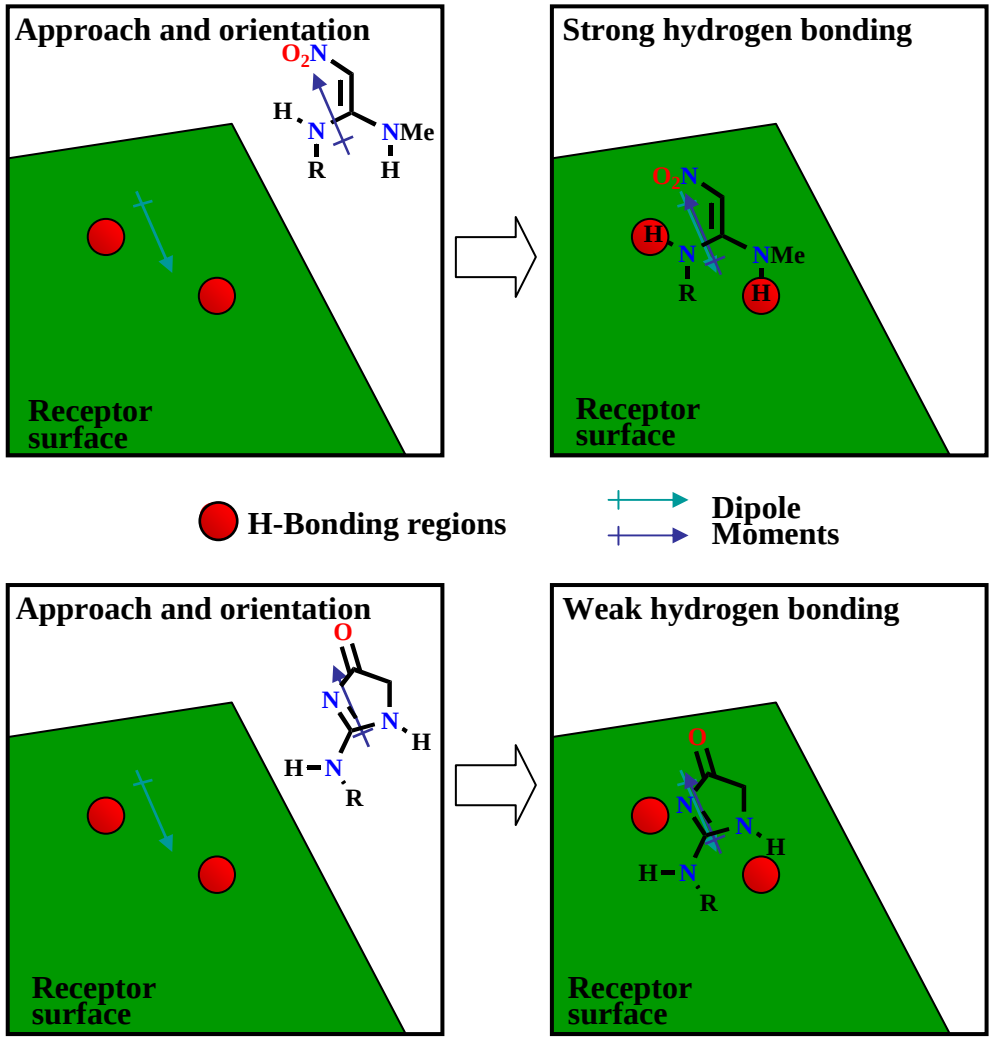
19. Dipole Moment Theory

19.1 Proposal -

- **A dipole-dipole interaction takes places between the drug and the binding site on approach of the drug**
- **The dipoles line up and orientate the drug**
- **Good interaction with the binding site occurs if the binding groups are positioned correctly w.r.t the binding regions - results in good activity**
- **Poor interaction occurs if the binding groups are not positioned correctly with respect to the binding regions - leads to poor activity**

19. Dipole Moment Theory

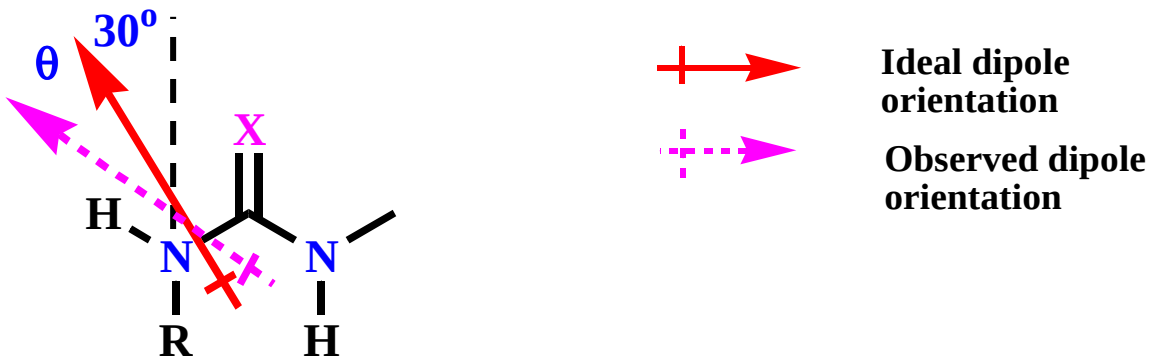
19.2 Dipole-dipole interactions



19. Dipole Moment Theory

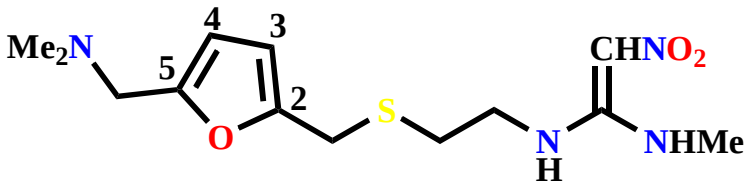
19.3 QSAR study including dipole-dipole interactions

- **The orientation of the dipole is more important than its strength**
- **Log (activity) = 9.12 cos θ + 0.6 log *P* -2.71**



- **Activity increases as hydrophobicity increases (log *P*)**
- **The ideal angle of the dipole moment = 30°**
- **At 30°, θ= 0° and cos θ = 1**
- **At 30°, Log (activity) = 9.12 + 0.6 log *P* - 2.71**
- **When dipole moment does not equal 30°, cos θ < 1 and activity falls**

20. Ranitidine (Zantac)



- **Contains a nitroketeneaminal group**
- **Different heterocyclic ring**
- **Took over from cimetidine as the most widely sold prescription drug in the world**

CFTII: backstage!