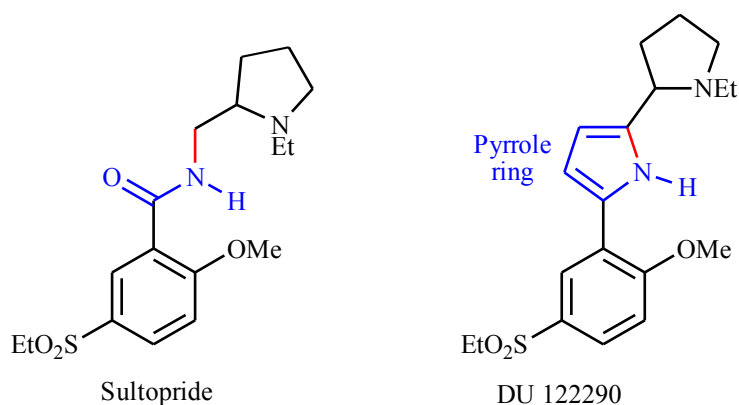
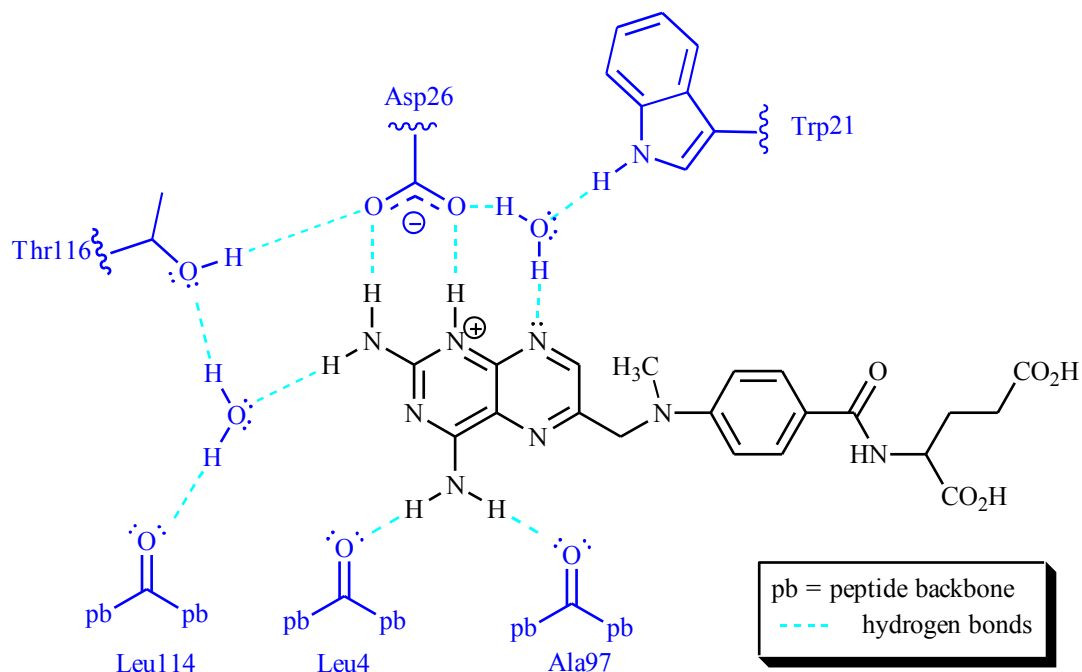


Answers to end-of-chapter questions

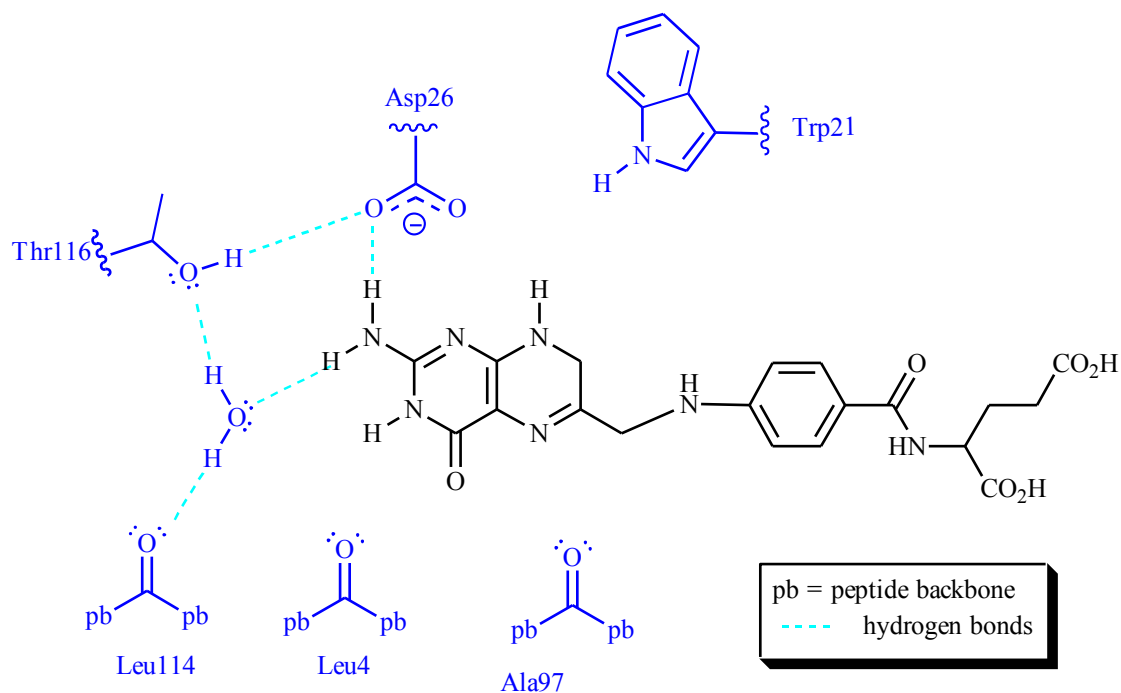
1) The pyrrole ring of DU 122290 serves to increase the rigidity of the side chain present in sultopride. The red coloured bond in sultopride is freely rotatable, but is locked within the pyrrole ring of DU 122290. This reduces the number of possible conformations that DU 122290 can adopt, and increases the chances of the active conformation being present when it enters the binding site, thus increasing activity. Moreover, some of the conformations that are now disallowed may be those which are adopted when sultopride binds to the D₂-receptor, thus increasing selectivity for the D₃-receptor.



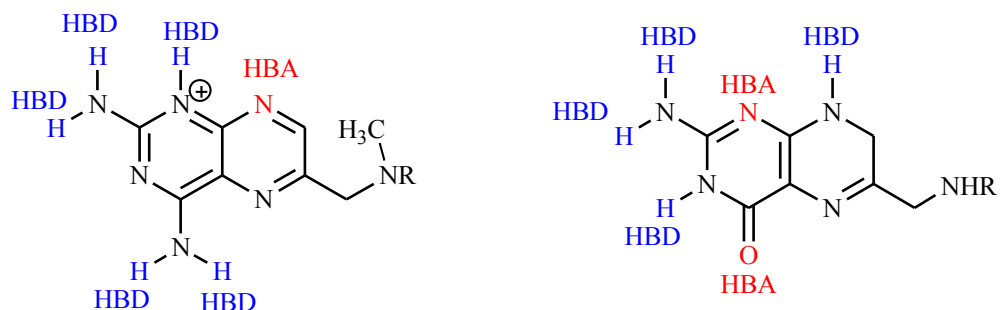
2) The binding interactions for methotrexate is shown below:



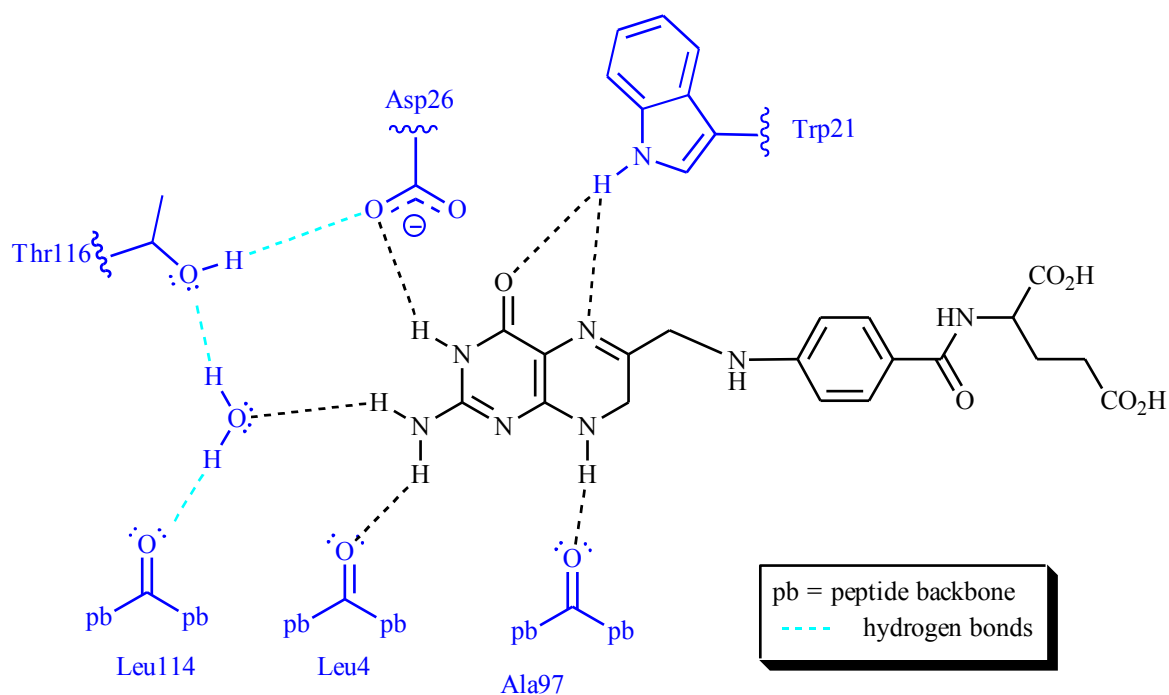
It is tempting to try and fit the natural substrate in the same manner. However, only the primary amino group interacting with Asp-26, and Leu-114 & Thr-116 (via a bridging water) can form the same interactions.



Comparing the two structures, there are important differences in the nature of the hydrogen bonding groups present.

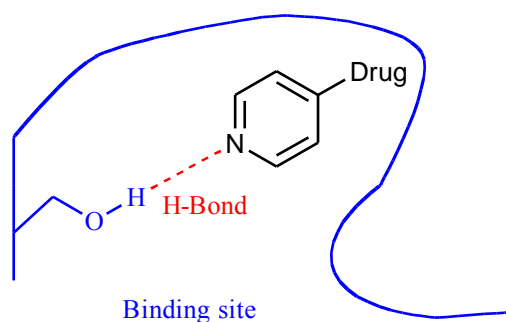


This suggests that different binding modes may be occurring here. For example, the binding mode shown below allows a greater number of interactions with the residues available in the binding site.

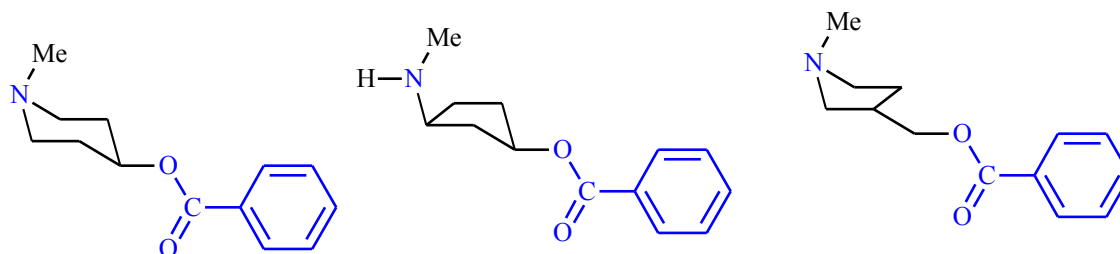


3) The results can be explained by proposing that the ester is acting as a prodrug. In the *in vivo* bioassays, the ester will be less polar than the carboxylic acid and so the prodrug will be able to cross fatty cell membranes such as those of the cells lining the gut wall. Once absorbed, esterases in the blood supply will hydrolyse the ester to reveal the carboxylic acid and generate the active drug. If the active drug is administered orally, the polar carboxylic acid hinders the drug crossing cell membranes and the drug fails to reach its target, accounting for the lack of activity. In the *in vitro* bioassay, the drug interacts directly with its target and is active, whereas the ester prodrug is inactive since the ester masks the important carboxylic acid group. There are no esterases present in the *in vitro* bioassay to hydrolyse the ester.

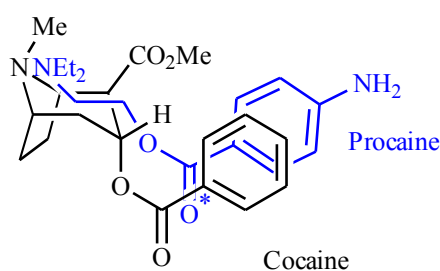
4) Structures I and II are similar in activity to the lead compound which demonstrates that replacing the aromatic ring with a pyridine ring has no significant effect in itself. The aromatic and pyridine rings can both interact with a binding site by van der Waals interactions. The increased activity for structure III strongly suggests that there is an additional binding interaction taking place with the binding site, such as a hydrogen bond to a serine residue. The strategy used here is ring variation, but could also be described as extension since an extra binding interaction has been discovered.



5) There are several possibilities. The following are three examples, with the first of these being the most obvious. All the structures lack the methyl ester since this is not part of the pharmacophore.

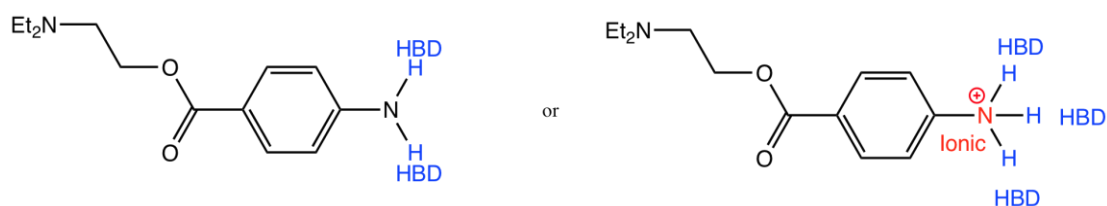


6) Cocaine is quite a rigid structure whereas procaine has a flexible chain with several rotatable bonds. As a result, it can adopt several conformations, one of which positions the important functional groups (amine, ester and aromatic ring) in the same relative position as in cocaine. This can be seen in the overlay below, where the alkyl chain of procaine bisects the ring of cocaine to place the amine group in the same position as that of cocaine.



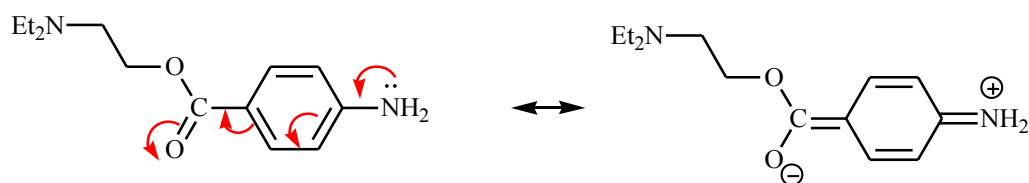
This is an exercise which can be carried out using a molecular modelling programme such as Chem3D or Spartan.

7) It could be interacting with an extra binding region in the binding site which is not used by cocaine. The interaction could be hydrogen bonding (HBD) and/or ionic bonding (if the amine is protonated). Note that the nitrogen of an aromatic amine is a poor HBA and is unlikely to be involved in a hydrogen bonding interaction.



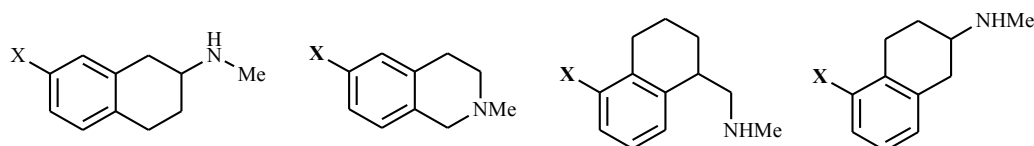
Modified diagram

An alternative explanation is that the amine could have an electronic influence on the *para* ester carbonyl group as shown below and increase its strength as a hydrogen bond acceptor.



Finally, it might not have any binding role at all and is present to increase the water solubility of the drug.

8) The idea of rigidification is to restrict the number of possible conformations that a molecule can adopt whilst retaining the active conformation. This means that the molecule is more likely to be in its active conformation when it interacts with its target, resulting in greater activity. Restricting the number of conformations also reduces the chances of side effects if conformations recognised by different targets are prevented. In this example, an extra ring can be added to lock bonds in the side chain such that they can no longer rotate. The following are examples. Many more are possible with different ring sizes and positions.

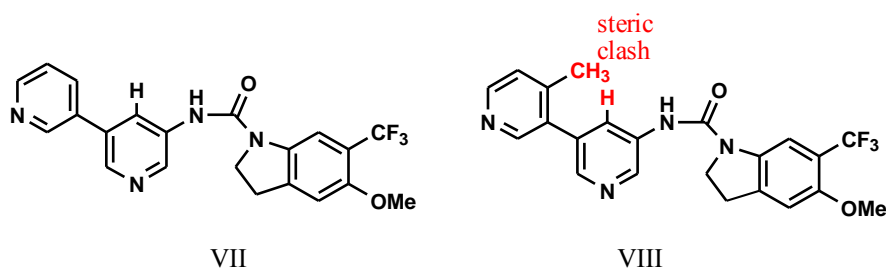


9) Rigidification. Inclusion of the alkene restricts the number of conformations in each analogue since no rotation is allowed round the double bond. The differences in activity between structures V and VI can be explained by proposing that the active conformation has been retained in structure V, but not in structure VI.

Structure V would have greater activity than the lead compound since there is more chance of the active conformation being present when the drug reaches the binding site.

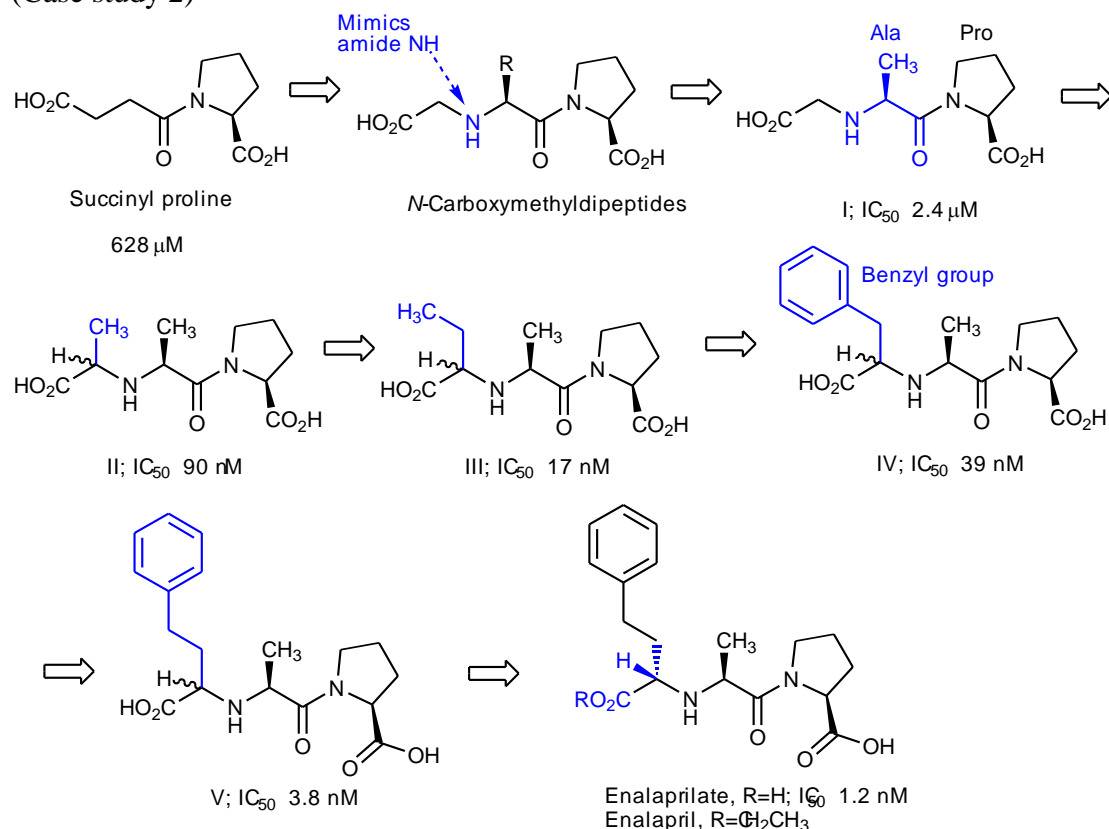
Structure VI might be expected to have no activity if the active conformation is disallowed. However, some activity may still be present if the molecule can fit the binding site in a different, but less efficient binding mode.

10) The methyl group in structure VIII is acting as a conformational blocker. It prevents the two pyridine rings becoming coplanar due to a bad steric interaction with a proton on the other ring.



If the activity increases, it suggests that the rings are not coplanar in the active conformation. Preventing the coplanar conformation increases the chances of the molecule being in the active conformation and increases activity (see also web article 5).

11) The design process leading to enalaprilate includes the structures shown below (Case study 2)



The lead compound is succinyl proline.

N-Carboxymethyldipeptides contain an NH group that can participate in hydrogen bonding and so this would be an example of an extension strategy where an extra functional group has been added to obtain an additional binding interaction.

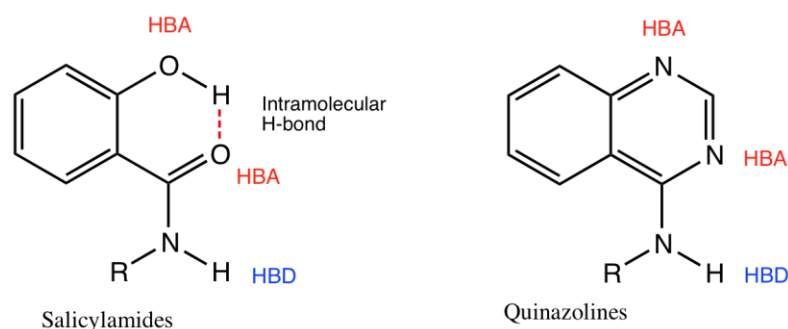
Structure I has a methyl side chain. This could be viewed as substituent variation as various R groups were tried out with the N-carboxymethyldipeptides. However, it

could also be viewed as an extension strategy as an additional binding interaction has been obtained where the methyl group forms additional van der Waals interactions with a hydrophobic pocket in the active site.

Structures II, III and IV are also examples of substituent variation where different substituents are tried out.

Structure V is an example of chain extension where the chain separating the aromatic ring from the main chain has been extended. The fact that activity increases suggests that the aromatic ring has been positioned more effectively to interact with a hydrophobic region in the active site.

12) Quinazolines could act as bio-isosteres for the salicylamides as they contain groups that are capable of matching the two HBAs and HBD required. Moreover, an intramolecular hydrogen bond is possible in salicylamides which would lock the phenol and amide substituents in the positions shown below – an example of rigidification. The quinazolines have a similar spatial arrangement of the key binding groups.



13) The analogue ($X=O$) lacks the hydrogen bonding interaction that is present for structure IX which explains the drop in binding affinity.

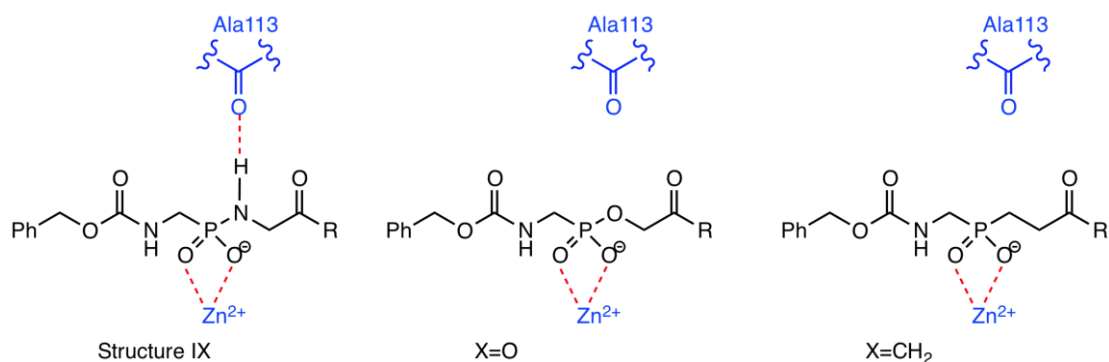
The analogue ($X=CH_2$) also lacks this hydrogen bonding interaction and so one might expect a similar drop in binding affinity for this structure. However, the binding affinity is roughly the same as for structure IX. This can be explained by considering desolvation energies. The NH group in structure IX is polar and is likely to be solvated before the inhibitor enters the active site, and so energy is required to remove that water of solvation before the group can form a hydrogen bonding interaction with the active site.

For the analogue ($X=CH_2$), the methylene group is hydrophobic and will not be solvated. Therefore, there is no energy penalty involved in removing water of solvation.

The overall binding affinity for a ligand will depend on

- the strength of favourable binding interactions achieved between the inhibitor and the active site and
- the energy expended in removing any water of solvation which will be detrimental.

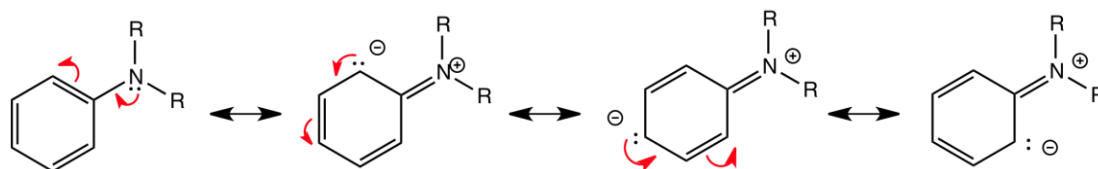
The fact that the analogue ($X=CH_2$) has a similar binding affinity to structure IX implies that the beneficial binding energy obtained from the extra hydrogen bond in structure IX is counterbalanced by the energy penalty required to remove water of solvation from around that group.



14) In all these cases the oxygen atom is next to an sp^2 centre. Therefore, it is possible for a lone pair of electrons on the oxygen atom to interact with the neighbouring pi system through resonance. As a result, it is less available to act as a hydrogen bond acceptor. Therefore, one would expect these oxygen atoms to act as weak hydrogen bond acceptors.

15) The first structure contains a pyrrole ring where the nitrogen's lone pair of electrons are part of an aromatic sextet. Therefore, the nitrogen will be a very poor hydrogen bond acceptor.

The second structure is an aromatic amine or aniline. In this case, the nitrogen's lone pair of electrons can interact with the aromatic system, and one can draw several resonance structures as below. Therefore, one would expect this nitrogen to be a poor hydrogen bond acceptor.



The third structure is a pyridine structure. Here, the nitrogen's lone pair of electrons is not part of an aromatic sextet and is unable to interact with a neighbouring pi system through resonance. Therefore, the nitrogen acts as a good hydrogen bond acceptor.