Chapter 3

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3.1 Initial concepts

At the start of chapter 3, we stated it was essential to have an understanding of isomerism to explain why:

- *The selective serotonin reuptake inhibitors (SSRIs) escitalopram and citalopram are used at different doses to treat depression*

Citalopram is a selective serotonin reuptake inhibitor commonly used in the treatment of depression. As we have outlined in chapter 3, citalopram contains a chiral centre and can therefore exist as two enantiomers: *R*-citalopram and *S*-citalopram. Like many drugs used in pharmacy, citalopram is, in practice, given as the racemate – this means that a tablet of citalopram contains an equal mixture of *R* and *S* enantiomers. Both enantiomers (as you would expect) bind to the serotonin transporter and inhibit the reuptake of serotonin. However, the *S*-enantiomer of citalopram has a much higher affinity for the serotonin transporter than the *R*-enantiomer, meaning that the *S*-enantiomer is responsible for eliciting much of the biological effect when citalopram is used. In view of this difference, pharmaceutical companies have formulated the *S*-enantiomer of citalopram as a single tablet for the treatment of depression. The *S*-enantiomer was given the name escitalopram (it's less confusing than using the term *S*-citalopram) and when compared to citalopram for the treatment of depression, due to the large differences in potency between enantiomers, escitalopram was found to be twice as potent than citalopram, meaning a 10 mg dose of escitalopram is equivalent to a 20 mg dose of citalopram (as the racemate). This difference offers escitalopram the potential advantage of reduced side effects and therefore an improved tolerability profile when compared to citalopram.

- *The antibiotic metronidazole interacts with the anti-coagulant agent warfarin (and thus increases the risk of a patient having a haemorrhage)*

As we have discussed for citalopram, warfarin also contains a chiral centre and can exist as two enantiomers: *S*-warfarin and *R*-warfarin. Like we have described for citalopram, above, warfarin is also used in practice as the racemate. Both enantiomers of warfarin have biological activity but the *S* enantiomer is known to be 3 – 5 times more active when compared to the *R* enantiomer. Each

enantiomer is also metabolised differently; for example, the enzyme, CYP 2C9, is responsible for metabolising the *S*-enantiomer of warfarin, while several enzymes, such as CYP 1A2 or CYP 3A4, are responsible for metabolising the *R*-enantiomer.

Metronidazole is an anaerobic antibiotic that works by targeting the DNA of bacteria. Metronidazole also inhibits the enzyme CYP 2C9 and thus, if used in combination with warfarin, it would inhibit the metabolism of the *S*-enantiomer, which, in turn, would increase the levels of S-warfarin levels in the body. As we know that S-warfarin is the more active enantiomer, the increase in concentration may cause the patient to have a serious bleed or haemorrhage.

- *Thalidomide – an anti-angiogenesis drug used in the treatment of multiple myeloma – is contraindicated in pregnancy.*

We discussed, in detail, in chapter 3 why thalidomide is contraindicated in pregnancy. If you can't remember why, then have a look at Integration Box 3.1 on page 66. In practice, if thalidomide is to be used in a woman of child-bearing age, it is very important to ensure she is not pregnant before the thalidomide is given. In many cases, the patient would have regular pregnancy tests throughout their treatment to ensure the patient does accidently become pregnant.

3.2 Conformations of linear molecules

In chapter 3, we looked at the possible conformations that a linear molecule can adopt by considering nabumetone as our example. We saw that this molecular prefers to adopt a staggered conformation around its -CH_2-CH_2- bond and that eclipsed conformations were of higher energy and therefore less favoured.

Besides the eclipsed and staggered descriptions, the other nomenclature used to describe the conformations is explained below:

- **Antiperiplanar**: a staggered conformation in which two particular atoms or groups on adjacent carbons are found at 180^o to each other;
- **Gauche** or **Synclinal**: staggered conformations with less than ±90^o between two particular atoms or groups on adjacent atoms;
- **Synperiplanar**: an eclipsed conformation in which in which two particular atoms or groups on adjacent atoms are aligned in the same plane, one behind the other;

- **Anticlinal**: eclipsed conformations with more than ±90^{o} between two particular atoms or groups on adjacent atoms.

Further examples

However, not all molecules, even simple ones, have such a clear preference for a staggered conformation. In this section, we will consider two simple examples in which the preferred conformation is not the one we would at first expect on the basis of a staggered conformation being most stable. This does not mean that our considerations of conformation so far are wrong; it means that there are other factors that can affect the conformational energy of particular molecules and that these can be so important that they 'override' the preference for the staggered antiperiplanar conformation we have seen with nabumetone.

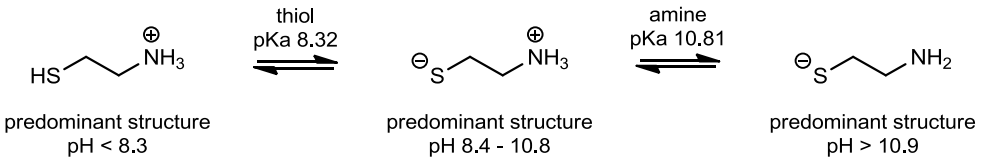
Example 1: cysteamine

Our first example is cysteamine, $NH_2CH_2CH_2SH$, the active agent for the treatment of a rare disease called cystinosis.

Did you know.....?

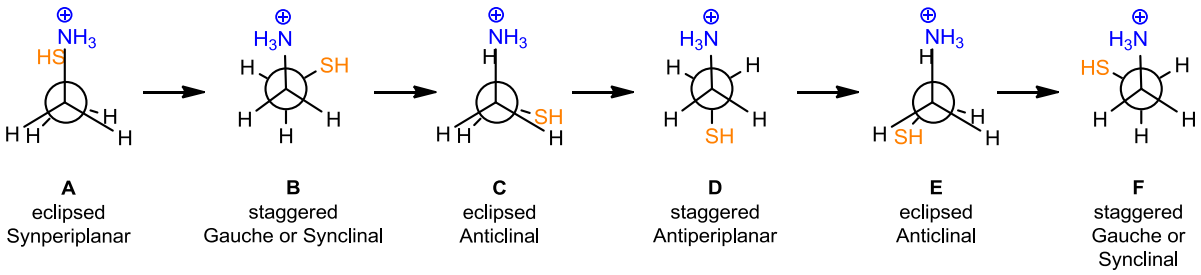
Cystinosis is a genetically inherited disease with an incidence in the UK, Europe and USA of approximately 1 in 150,000 live births, putting it into the category of a rare or ***orphan disease***. There are patients in other countries, but they are often not diagnosed or are even rarer; currently, there are only about 2,000 patients known worldwide. Cystinosis usually manifests during the 1st year of life as excessive thirst and urination, and is often at first mistaken for diabetes, then progresses to failure to thrive and renal Fanconi syndrome – a disease of the kidneys, in which reabsorption of essential electrolytes and other species, such as phosphate, bicarbonate, glucose, amino acids and uric acid, is poor. Cystinotic patients have a mutation or deletion in the gene for a transport protein, cystinosin, which is responsible for removing excess cystine from the lysosomes of cells. This leads to accumulation of cystine, a product from the breakdown of proteins, which crystallizes and eventually causes cell death. If untreated, a cystinotic child rarely survives beyond the age of 10 years. Fortunately, cysteamine was found to deplete the excess lysosomal cystine and was licensed for the treatment of cystinosis in the 1980s; if taken every six hours, it helps to prevent cell damage, although it remains common for cystinotic patients to require a kidney transplant. It is formulated as its bitartrate salt, Cystagon®. The main disadvantage to cysteamine is that it smells and tastes very unpleasant, even in the salt form, so researchers are looking into improving this treatment.

There are two methylene (CH_2) carbon atoms in cysteamine, one with an amine (NH_2) substituent and the other with a thiol (SH) group. Before we draw the conformations, we need to remember that amines usually have basic properties and thiol groups have weakly acidic properties, and we need to know the pKa values of the groups and the pH at which the molecule will be found. The pKa values of the amine and thiol in cysteamine are 10.81 and 8.32, respectively. For this discussion, we will consider cysteamine at pH 7.2, similar to that in the blood and cells of a patient. At pH 7.2, we can expect the amine to be almost 99.95% ionised and the thiol to be about 90% unionised, so the structure that will predominate can be represented as $H_3N^+CH_2CH_2SH$, Scheme 3.1.

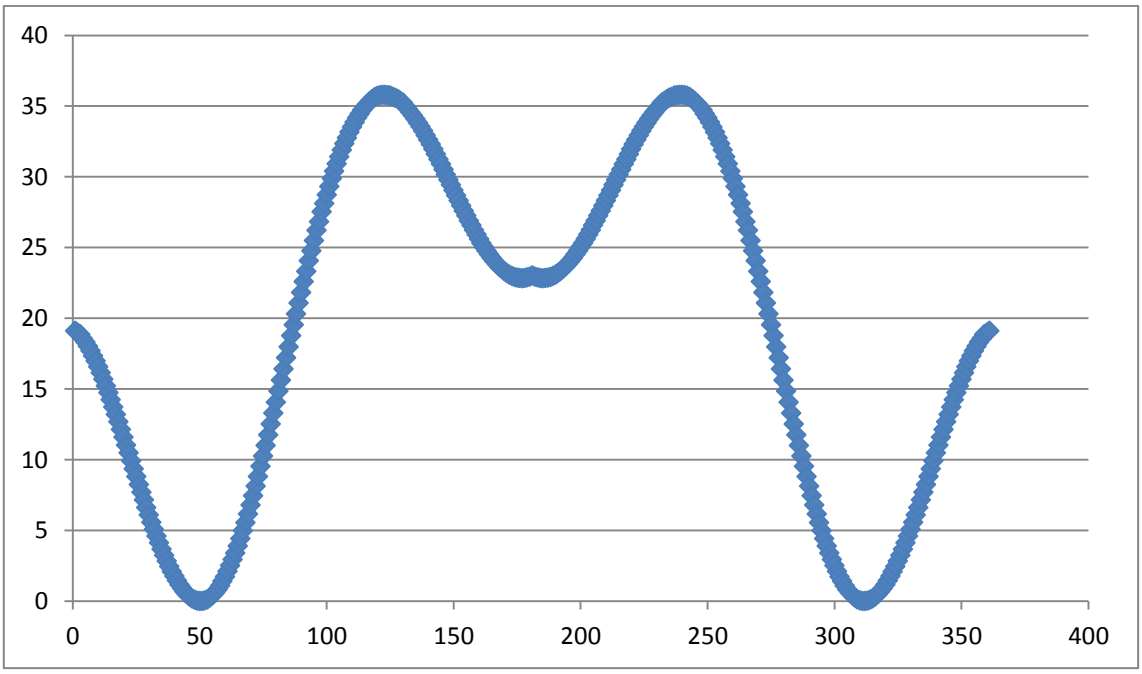


Scheme 3.1 Acid-base equilibria of cysteamine.

We can again use a Newman projection to look along the C-C bond and consider the possible conformations cysteamine can adopt. If we start with the two substituents arranged one behind the other, we have our starting point, conformation **A**; then, keeping the front carbon and its attached atoms / groups stationary and rotating the rear carbon atom clockwise around the C-C bond, we can trace the energy changes in this small molecule as it passes through the high and low energy conformations, Scheme 3.2 and Graph 3.2.



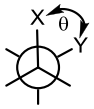
Scheme 3.2 Key conformations around the substituted two carbon unit of cysteamine.



Graph 3.2 Plot of energy changes as a function of dihedral angle[1] calculated at every whole degree between 0-360° for cysteamine at pH 7.2. *[Calculations performed using Gaussian by Peter Dawson.]*

When we look at the energy of each conformation in Graph 3.2, we find that the plot of energy against dihedral angle is quite different to the plot we saw for nabumetone. Why is this plot so different? Well, the possibility for hydrogen bonding of the ionised amine and the thiol group to each other, or through a water molecule, means that other factors influence which conformation is preferred more than the intramolecular force of torsional strain. We can use computer-based molecular modelling programmes, such as Gaussian, to calculate the most likely conformations of cysteamine. Two possible conformations that are consistent with the preferred energy calculations are shown below. Each possibility is shown from the side and as a Newman projection. Structure **G** shows a hydrogen bond directly between the thiol and protonated amine, while structure **H** uses a water molecule to mediate the hydrogen bond between the same groups. In both cases, the structure is held in a gauche conformation to get the best possible hydrogen bond. Other hydrogen

[1] The dihedral angle between two substituents, X and Y, is the angle between the two planes in which the substituents lie and is described by θ (theta).



bonded possibilities exist according to molecular modelling calculations, but the gauche version is consistently found to lie at an energy minimum.

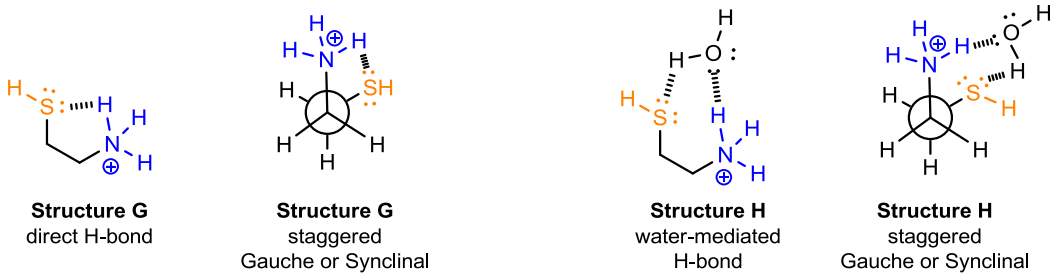
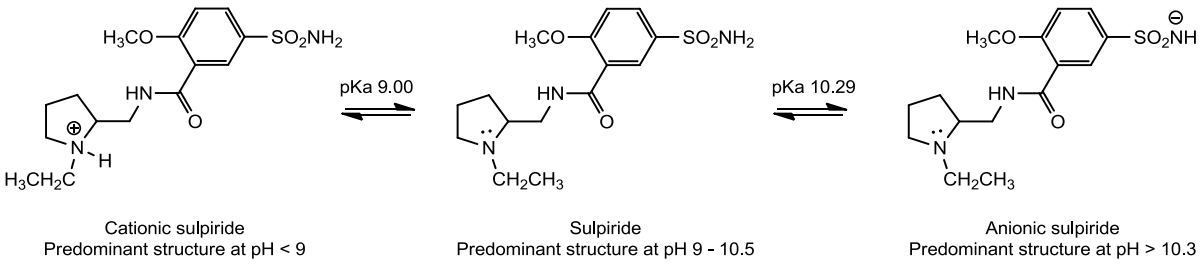


Figure 3.1 Possible hydrogen bonding stabilisation of the gauche conformation of cysteamine

Example 2: sulpiride

Our second example is an antipsychotic agent named sulpiride, in which intramolecular hydrogen bonding has been proposed to favour the conformation required to bind to its pharmacological target, the dopamine D_2 receptor, at which it shows antagonist activity. Sulpiride has two pKa values: 9.00 and 10.29. The first is the pKa of the basic tertiary amine group and the second value represents the acidic sulfonamide group; the ionised amine is likely to be most prevalent in the blood, while the sulfonamide group does not ionise significantly at physiological pH, scheme 3.3.



Scheme 3.3 Acid-base equilibria of sulpiride.

The conformation of cationic sulpiride was calculated by molecular modelling and measured by spectroscopic methods. The most stable conformation was found to involve an intramolecular hydrogen bond from the ionised tertiary amine to the carbonyl of the amide group, which promotes the stability of an eclipsed form suggested to be the required conformation for binding to the dopamine D_2 receptor, Figure 3.2.

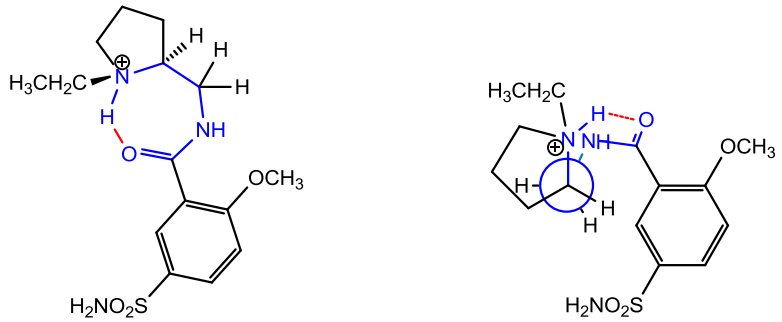


Figure 3.2 Intramolecular hydrogen bonding in sulpiride favours the eclipsed conformation.

3.3 Conformations of cyclic molecules

In chapter 3 we looked briefly at the conformations of cyclic molecules and found that, as for acyclic molecules, conformations that allow staggering of bonds on adjacent atoms are favoured; however, there are fewer conformations possible for cyclic structures. We considered saturated six-membered rings and found that the chair form is preferred over the boat form, mostly due to torsion and steric strain in the boat form. When a molecule has only one six-membered ring, it can interconvert between chair and boat forms to explore the possibilities for its restricted conformations; when there are two or more rings fused together, ring flip is more difficult and frequently not possible. How does a cyclohexane-based molecule flip its conformation from chair to boat and from chair to chair forms? You can follow the interconversion of cyclic conformations in Figures 3.3, 3.4 and 3.5 below, which show how one chair form converts to the other chair form by passing through a boat form for *trans*- and *cis*-1,2-, 1,3-, and 1,4-disubstituted cyclohexanes, respectively. If you follow the substituents X and Y in each diagram, you can see how a substituent starting in the equatorial position finishes in the axial position on the opposite chair form, while an axial substituent on the initial chair form ends up as equatorial on the opposite chair form. To understand fully, it is recommended that you make a model and try it for yourself.

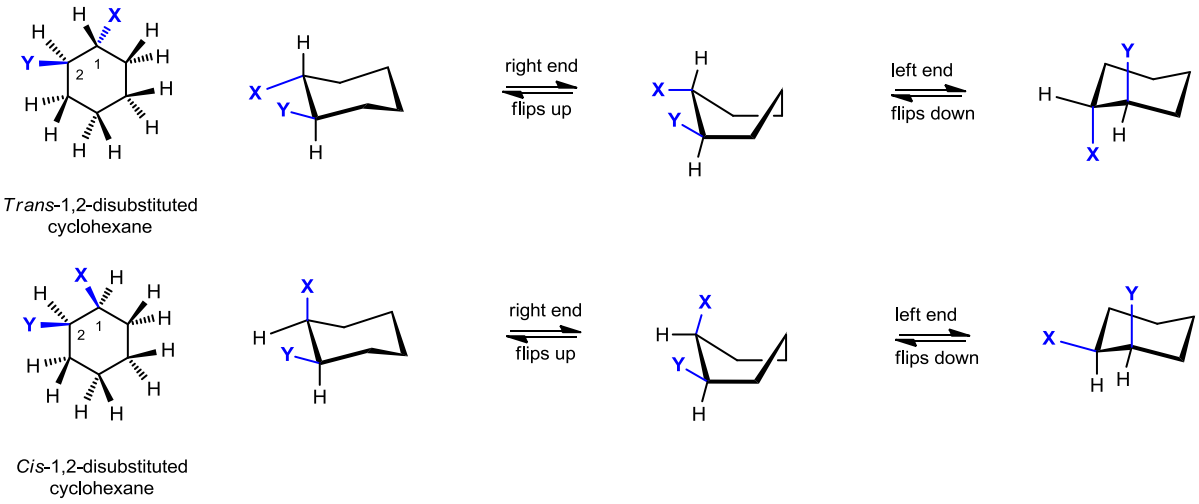


Figure 3.3 Interconversion of *trans*- and *cis*-1,2-disubstituted cyclohexane chair and boat conformations.

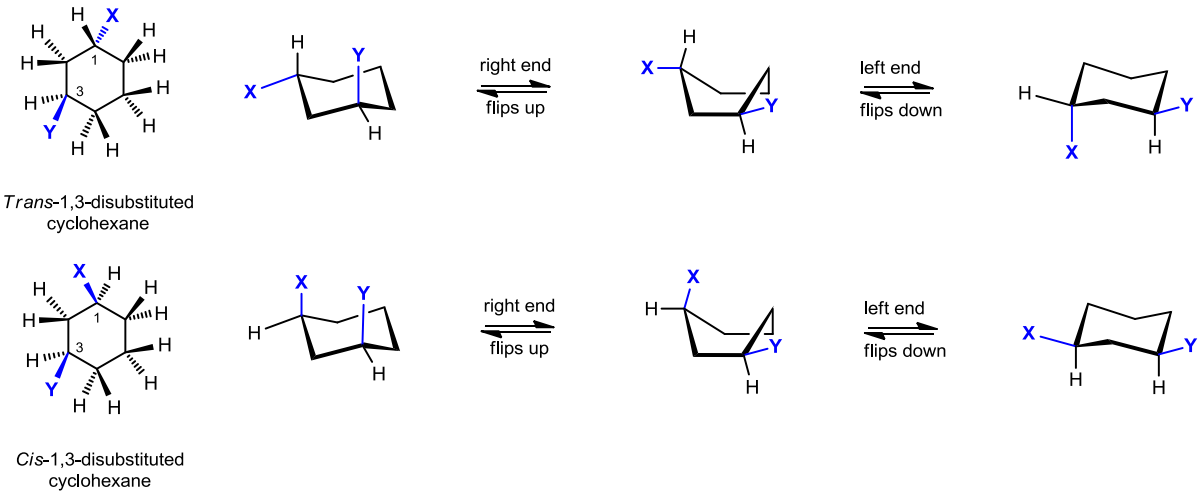


Figure 3.4 Interconversion of *trans*- and *cis*-1,3-disubstituted cyclohexane chair and boat conformations.

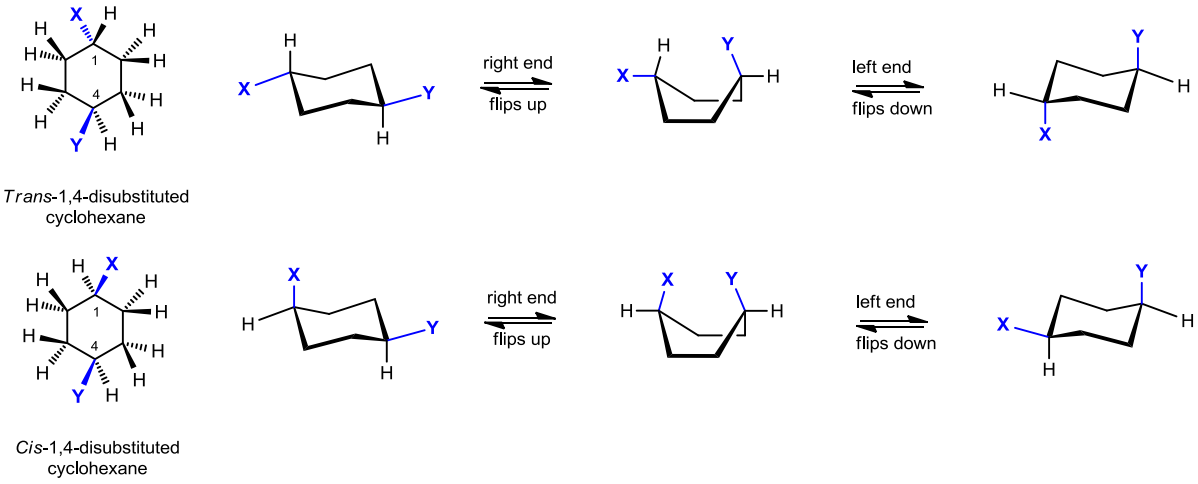
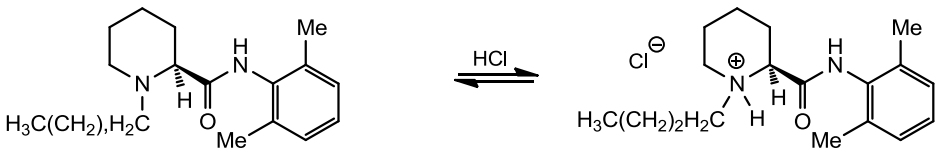


Figure 3.5 Interconversion of *trans*- and *cis*-1,4-disubstituted cyclohexane chair and boat conformations.

You can see from these conformations that the *cis*-1,4-disubstituted boat form, with the X and Y substituents on the flagpole positions and on the same face, is particularly disfavoured, due to high steric strain associated with the close spatial position of these groups. Note that the *cis*- (on the same face) and *trans*- (on opposite faces) terminology is still used for these cyclic molecules; there is little opportunity for confusion with these systems.

By considering the most stable conformations of cyclic molecules, you can work out which are the conformations likely to be favoured. Take levobupivacaine hydrochloride for example; this is a local anaesthetic. The first thing is to identify the ionisable nitrogen atom that becomes protonated to form the hydrochloride salt: the tertiary amine. Remember that amide nitrogen atoms are not basic as their lone pair of electrons is involved in resonance with the adjoining carbonyl group and is not available to accept an H^+, Figure 3.6.



Levobupivacaine Levobupivacaine hydrochloride

Figure 3.6 Levobupivacaine and its hydrochloride salt: the active local anaesthetic.

Once protonated, you can see that levobupivacaine hydrochloride is similar to the 1,2-disubstituted cyclohexane example in Figure 3.3. The stereochemistry at the chiral carbon is fixed (as *S*), but amines are able to invert; which of the conformations in Figure 3.7 do you think will be preferred?

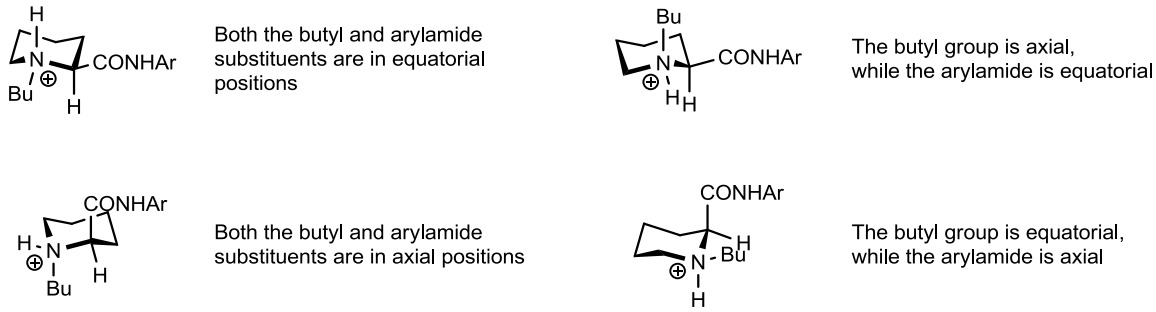


Figure 3.7 Possible conformations of ionised levobupivacaine.

You are correct if you saw that one structure has both substituents in the equatorial position, which minimises all strain and is likely to be preferred. When any of the substituents are in the axial position, the structure suffers from steric strain due to the close proximity in space of the other axial atoms in this conformation.

Tranexamic acid provides an example of a 1,4-disubstituted cyclohexane; this is an anti-fibrinolytic agent, which is used clinically to control bleeding. In the body, clot formation is prevented by the breakdown of fibrin by plasmin; tranexamic acid inhibits the action of plasmin on fibrin and so allows clots to form and reduces bleeding. The substituents occupy the 1 and 4 positions and have a *trans*-relationship to each other: one is above the plane of the ring, while the other is sited below. This allows both substituents to be equatorial in one of the possible chair conformations, Figure 3.8.

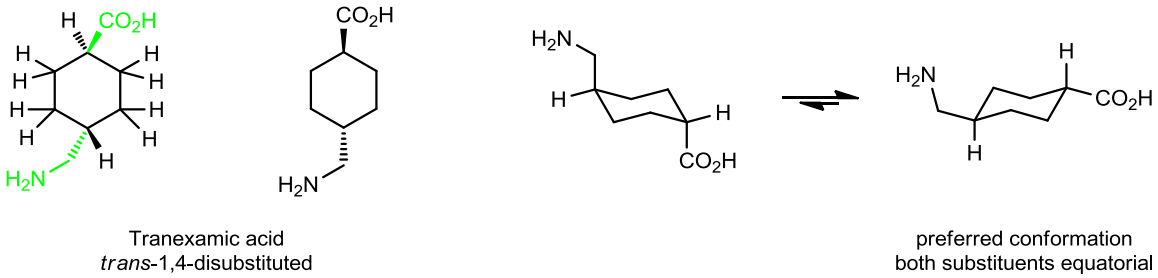


Figure 3.8 Tranexamic acid: the conformation with both substituents equatorial is preferred.

Saturated five-membered ring systems are also common in pharmaceuticals and usually adopt a defined ‘envelope’ or ‘twist’ conformation. For example, lamivudine is an anti-viral agent that is used clinically to treat HIV and hepatitis B virus; it acts by inhibiting reverse transcriptase, a viral enzyme

that uses viral RNA to make the corresponding viral DNA for incorporation into the host cell's DNA. The crystal structure of this agent shows that its five-membered oxathiolane ring adopts an envelope conformation, Figure 3.9.

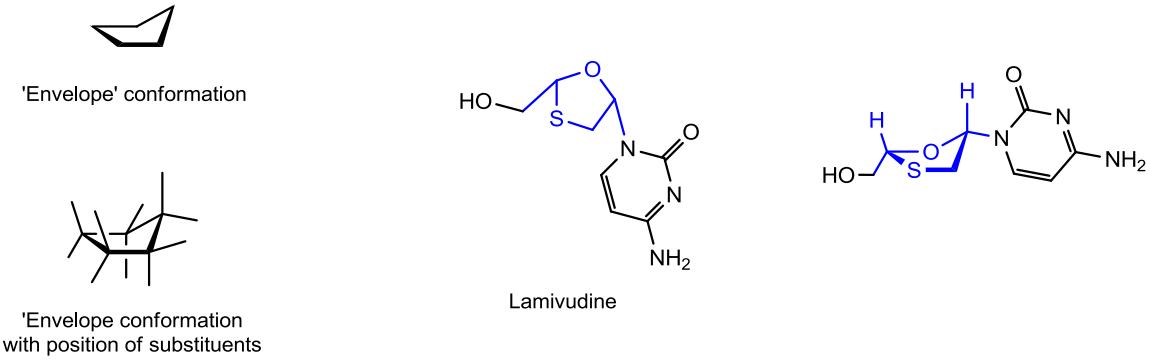


Figure 3.9 The anti-viral agent, lamivudine, adopts an envelope conformation for its five-membered oxathiolane ring.

The twist structure is created by three consecutive atoms in the same plane, with the remaining two atoms from this cyclic system, as the name suggests, twisted out of the plane, one above and one below. Lactulose is a laxative, based on the structure of the natural sugar, fructose. It has a 6 membered hexose ring that adopts a chair conformation, with most of the substituents in the equatorial position, and a 5 membered furanose ring that adopts a twist form, which is very common for furanose structures, Figure 3.10.

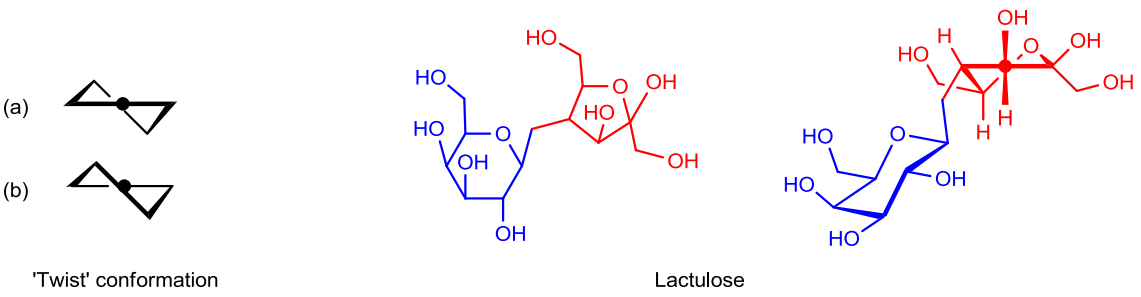


Figure 3.10 The common twist conformation of furanose is seen in lactulose; the solid sphere in the long bond indicates the location of the central atom of the three in-plane atoms, (a) three in-plane atoms at the front and two out of plane atoms at the back; (b) three in-plane atoms at the back and two out of plane atoms at the front.

Four-membered and three-membered rings are very strained and have little conformational options available to them. Four-membered rings try to adopt an open V-shape when possible to minimise

the number of adjacent bonds in the same plane (remember, adjacent bonds in the same plane gives rise to torsional strain). Three-membered rings can only be planar. The structures of tranylcypromine, an antidepressant that acts as a monoamine oxidase inhibitor, and sibutramine, a molecule with appetite suppressant activity (now withdrawn from the UK market), are shown as examples, Figure 3.11. The cyclobutyl conformations can invert, in a similar to manner to the cyclohexyl conformational interconversions we have already seen.

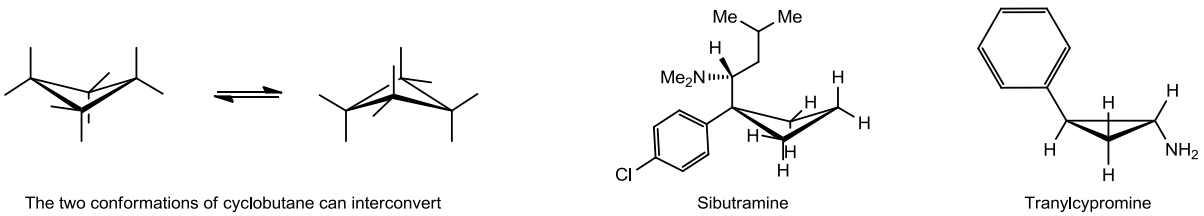


Figure 3.11 The conformations of tranylcypromine and carboplatin; note that the substituents of sibutramine are not shown to scale.

These cyclic systems try to adopt the same preferred shapes even when they are fused together. There are many examples of pharmaceutical products with fused cyclic systems; some of these are shown below. Beta-lactam antibiotics have a fused 4-membered and 5-membered bicyclic system; granisetron, an anti-emetic agent, has two fused 6-membered rings; hyoscyamine is a plant-derived anticholinergic / anti-spasmodic agent that has been used to relieve gastrointestinal disorders, such as irritable bowel syndrome and diverticulitis, and consists of five-membered and six-membered rings fused together; steroids, such as cholesterol and cholic acid (a bile salt), typically have three 6 membered rings fused to a 5 membered ring, Figure 3.12.

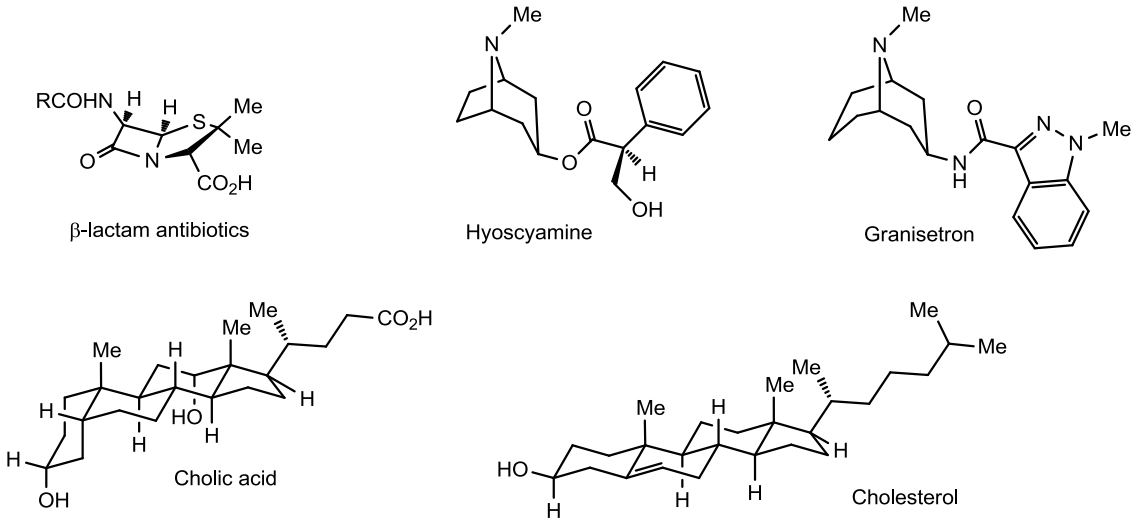
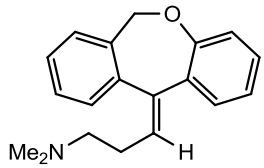


Figure 3.12 Examples of fused ring systems.

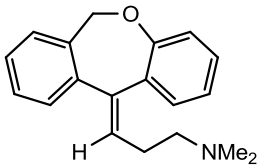
3.4 Optical isomers

Practice in assigning E and Z to alkenes

Doxepin is a tricyclic antidepressant used in the treatment of major depression and insomnia; it also has sedative activity. The alkene between the dibenzoxepine (tricyclic) core and the ionisable amine side chain gives rise to geometric *Z* and *E* isomers. Doxepin is administered as a mixture of the isomers in a ratio of 15 *Z* : 85 *E*. The *E*-isomer has greater antidepressant activity through its inhibition of serotonin reuptake by pre-synaptic neurons, while the *Z*-isomer has greater activity as an antagonist of histamine H1 and H2 receptors. Consider the structures in Figure 3.13 and make sure you can assign the geometry correctly. The pharmacological activity of each will help you to check your answer with the statement above.



Doxepin: isomer with greater antidepressant activity



Doxepin: isomer with greater sedative activity

Figure 3.13 Doxepin geometric isomers

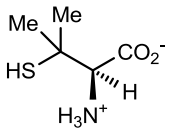
Practice in assigning chirality

Some examples follow for you to practice your skills in assigning stereochemistry, with answers in Figure 3.17. We will also meet some additional nomenclature that is less commonly used now, but which you may meet at some time.

A term that you may see used in relation to pharmaceutical enantiomers is the **eudysmic (or eudismic) ratio**, which describes the differences in pharmacological activity between two drug

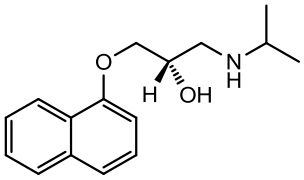
enantiomers. The active form with the desired pharmacological activity is called the **eutomer**, while the enantiomer, with either undesirable or absent biological effects is called the **distomer**.

Penicillamine is active in slowing the progression of rheumatoid arthritis and is also used to treat Wilson's disease, in which a genetic defect makes the body unable to metabolise copper, leading to its accumulation in the tissues and causing neurological disease. Only one enantiomer possesses the ability to chelate the excess copper and remove it from cells, while the other enantiomer is toxic and can cause blindness. Obviously, it is vitally important to ensure that patients receive the correct enantiomer; assign the stereochemistry to find out which is the eutomer.



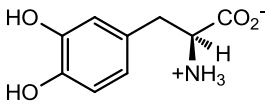
Penicillamine

Propranolol is an example of a beta-blocker, which blocks the beta-adrenoceptors in the heart and other tissues. It acts to relieve hypertension and helps to prevent angina, myocardial infarction (heart attack) and arrhythmias. Due to the lack of adverse effects of the distomer, propranolol is administered as the racemate; assign the stereochemistry to the structure shown here to identify the eutomer.



Propranolol

Levodopa is the natural amino acid precursor of dopamine, an important neurotransmitter in the brain. The eutomer is administered in its enantiomerically pure form to treat Parkinson's disease, as the distomer causes granulocytopenia and other serious side effects. The eutomer is shown here; assign the stereochemistry to identify which enantiomer is the active drug.



Levodopa

Many pharmaceuticals have more than one chiral carbon atom, giving rise to several possible diastereoisomers with the activity usually residing in only one form. If the other possible diastereoisomers have no adverse effects, the administered drug may be a mixture of them all; however, more usually, a pair of enantiomers would be administered. The pair of enantiomers is sometimes labelled as the ***threo***-form or the ***erythro***-form. Let's take an example of a drug with two chiral carbon atoms to illustrate this concept: we met ephedrine and pseudoephedrine in chapter 3. If you have completed the self-check activity 3.10, you will have found that ephedrine has *1R, 2S* stereochemistry and pseudoephedrine has *1S, 2S* stereochemistry. If we draw out the 4 possible diastereoisomers as the two pairs of enantiomers in a slightly different way, we see that ephedrine

and its enantiomer have the number 1 priority groups OH and NHMe (according to the Cahn-Ingold-Prelog system) on the same side, while pseudoephedrine and its enantiomer have the number 1 priority groups on opposite sides, Figure 3.14. When a molecule with two chiral carbon atoms has the two priority 1 groups on the same side, it is given the prefix ***erythro***; if they are on opposite sides, the prefix ***threo*** is used. You will sometimes see ephedrine labelled as *erythro*-ephedrine and pseudoephedrine labelled as *threo*-ephedrine.

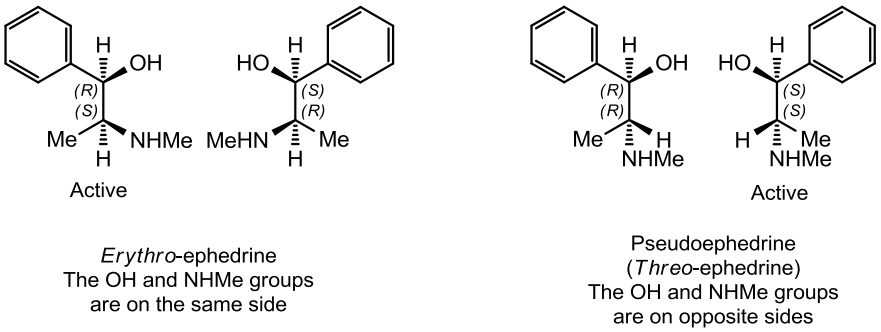


Figure 3.14 The *erythro*- and *threo*-forms of ephedrine.

Ephedrine allows us to meet another term used in describing some chiral molecules: **epimers**. Epimers are diastereoisomers that differ at only one possible chiral centre. The two active forms of ephedrine are (*1R, 2S*)-ephedrine and (*1S, 2S*)-ephedrine, also called pseudoephedrine. C2 in both molecules has *S* stereochemistry and only C1 varies, so these molecules are also termed epimers. Sugars provide further examples of molecules that differ at only one possible chiral centre, for example α-D-glucopyranose and β-D-glucopyranose, Figure 3.15: these molecules each have 5 chiral carbons, yet only differ at C1, making them epimers. You will have learned all about these molecules in chapter 9.

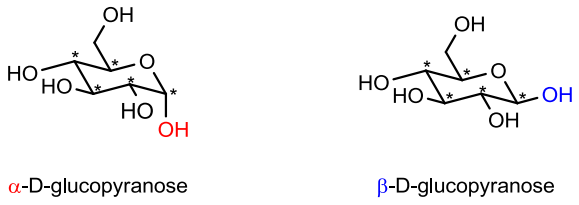
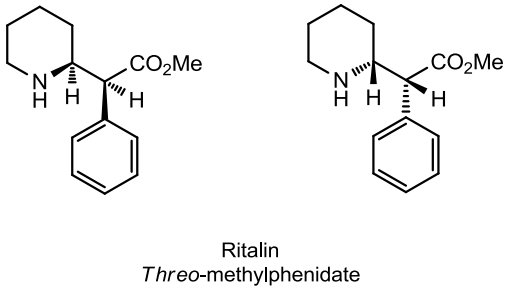


Figure 3.15 The epimers of D-glucopyranose

Ritalin provides an example of a pharmaceutical administered as a pair of 2 enantiomers out of 4 possible diastereoisomers: it is used to treat ADHD (attention deficit hyperactivity disorder). Ritalin has two chiral carbon atoms and is administered as the racemic mixture of the *R,R* and *S,S* forms

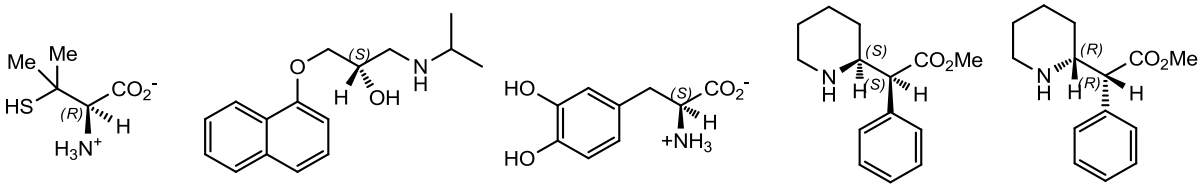
(also called the *threo*-form). Only the *S,S*-enantiomer has the CNS stimulant activity required to treat ADHD; it is believed that the *R,R*-enantiomer has no significant activity and no adverse effects. Assign the stereochemistry to the two structures in Figure 3.16 and decide which is the active S,S-enantiomer.



Ritalin
Threo-methylphenidate

Figure 3.16 The *threo*-enantiomers of methylphenidate (Ritalin)

The answers to these stereochemistry assignments are as follows:

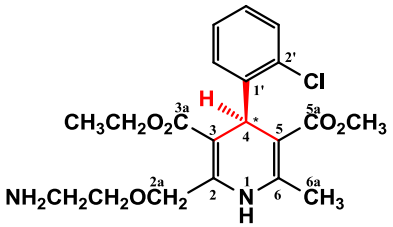


Penicillamine | Propranolol | Levodopa | Ritalin *Threo*-methylphenidate

Figure 3.17 Answers to chirality / stereochemistry examples.

Assigning absolute chirality in cyclic systems

In the case of cyclic molecules, the same rules are followed, as shown in the following example.



Amlodipine

Amlodipine is currently marketed as the racemic mixture, but one enantiomer shows about 1000x greater calcium channel blocking activity (for the treatment of hypertension and angina).

The structure of the eutomer (active enantiomer) is shown here; we can use the rules above to assign it correctly as *R* or *S*.

Follow the steps:

1. Identify the chiral carbon atom (here it is indicated by a *);
2. Make sure it is drawn as a 3D carbon atom (i.e. make sure it has wedge and hashed bond to show 3D stereochemistry);
3. Identify and prioritise the atoms around the chiral carbon atom:
    a. H (priority 4), C3, C5 and C1'. C3 has three bonds to C;
    b. C5 has three bonds to C; C1' also has three bonds to C.

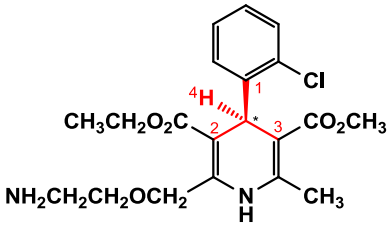
    Moving further afield,

    c. C3a has 3 bonds to O,
    d. C5a has 3 bonds to O,
    e. C2' has 2 bonds to C and 1 to Cl;

    From this, we can see that **C1' takes priority 1**, as it has an atom with a higher atomic number (Cl, *Z* = 17) sited 2 bonds away.

    We still need to decide whether C3 or C5 takes priority 2:

    f. moving to C2 and C6, both have the same atoms, C and N, attached to them;
    g. however, when we get to C2a and C6a, we find that one of them (C2a) has 2 bonds to H and 1 to O, while the other (C6a) has 3 bonds to H,
    h. this means that **C3 takes priority over C5**, as it has higher priority atoms attached sequentially to it.
4. Adding the priority to the four atoms attached to C4, we get:
5. Check that the priority 4 atom is now at the back, on the end of the hashed bond, and then ignore it.



Look at the order 1,2,3. In this case, it goes anticlockwise, so the chiral carbon is given the designation *S*.

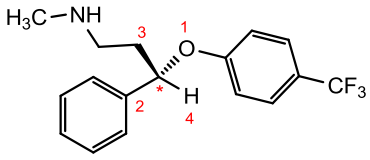
Rotating structures to put the lowest priority group at the back

All of the examples we have considered so far have been kindly drawn with the number 4 priority group at the back; however, that is not always the case. In chapter 3, we showed you a method of

looking along the C-H bond (or the bond from the chiral C to the number 4 group); however, this can be difficult with some structures. Alternatively, to assign the stereochemistry reliably, the structure should be redrawn to put the number 4 priority group at the back. This must be done with great care, or you could invert the stereochemistry. Here we show you a way of redrawing the structure that will reliably maintain the correct stereochemistry and allow you to assign it; for comparison, we will use the same example as we used in chapter 3: fluoxetine.

HINT: It is easier to redraw a chiral carbon correctly if one of the bonds in the plane of the paper is kept still and the rest of the molecule is rotated around this one stationary bond.

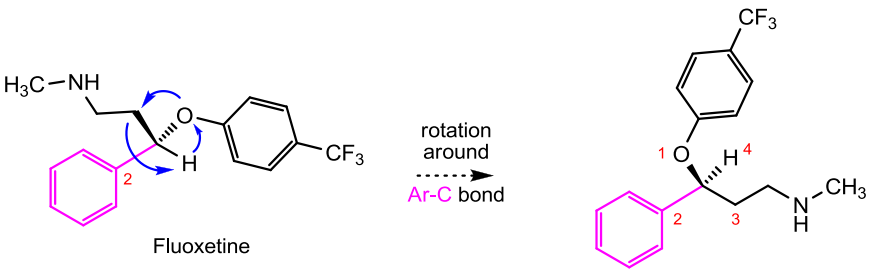
Remember fluoxetine is a serotonin selective reuptake inhibitor (SSRI) marketed as Prozac®, sometimes used in the treatment of depression; the more potent enantiomer is shown below:



Fluoxetine

After identifying the chiral carbon, you can assign priority to the groups around it. The H atom is the priority 4 group, but it is not behind the plane of the paper. If you try to assign the chirality in this representation of fluoxetine with the H in the plane of the paper, you will find that it looks like *R* stereochemistry, but **this is incorrect**.

To correctly assign the stereochemistry with certainty, the **H atom must not lie in the plane** of the paper. To redraw the molecule with the H atom at the back, we can keep the phenyl ring (priority group 2) stationary and imagine turning the remaining three groups like the steering wheel of a car. As you imagine the rotation, you can move the H to the back to replace the OAr group behind the plane of the paper, the OAr group moves to the position in front of the plane of the paper and the alkyl amine moves to the place where the H started — in the plane of the paper, Figure 3.18.



Fluoxetine

(*S*)-Fluoxetine

Figure 3.18 Redrawing of fluoxetine to correctly identify the chirality.

Once the H atom is at the back, we can immediately see that the priority 1 to 2 to 3 follows an anticlockwise direction, meaning that **(*S*)-fluoxetine is the active enantiomer**.

Atropisomerism

In chapter 3, we briefly looked at atropisomerism, which occurs in certain biphenyl molecules when they are asymmetrically substituted in the 2,6 and 2',6' positions by atoms large enough to prevent free rotation around the bond joining the aromatic rings (CH_3, OH, NH_2 are sufficiently large). In these systems, the two aromatic systems lie orthogonal to each other and when A ≠ B and A' ≠ B', then stereoisomerism arises, Figure 3.19.

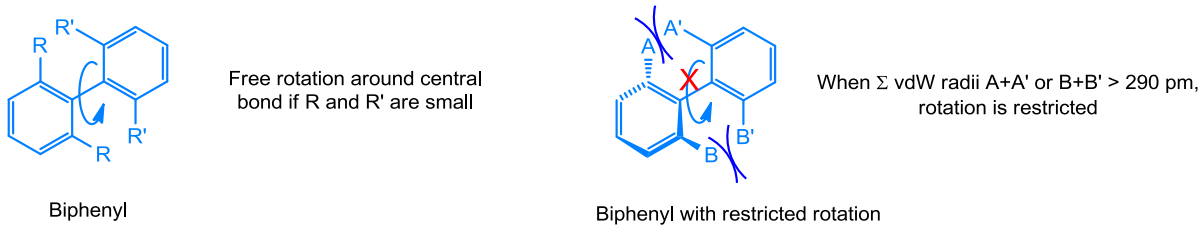


Figure 3.19 Atropisomerism of a biphenyl system due to restricted rotation around the central bond.

We considered the case of gossypol, a natural phenolic binaphthaldehyde isolated from the cotton plant *Gossypium*. It has been shown to possess male anti-fertility, antimalarial and anticancer properties. In gossypol, A is a phenol (OH) group and B is a methyl (CH_3) group. Restricted rotation around the central biphenyl bond results, generating enantiomers of gossypol, Figure 3.20.

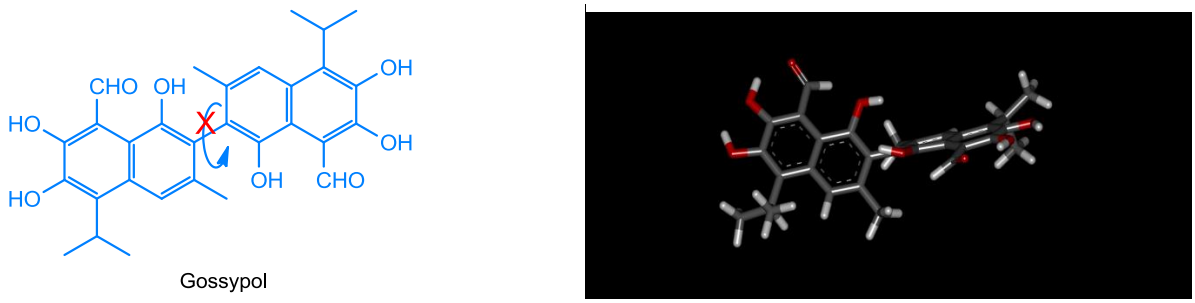


Figure 3.20 Gossypol, a natural molecule that exhibits atropisomerism.

Notice how our 2D structural representation on the left does not show the orthogonal relationship of the two naphthyl systems; this becomes apparent in the picture on the right, in which you can

clearly see the 3D structure. Beyond causing asymmetry in the system, this orthogonal relationship also prevents delocalisation of the electrons across the two naphthyl systems, as delocalised systems must be co-planar.

The assignment of stereochemistry of atropisomers follows the same Cahn-Ingold-Prelog rules that we have already used, but is rarely required; however, the rules are explained below using gossypol as the example.

Did you know.....

Gossypol has been the subject of considerable study, sponsored by the World Health Organisation, as an orally active male antifertility agent and as an anticancer agent. It occurs naturally in cotton seed oil and its male antifertility activity was noted in China, where the oil was sometimes used for cooking. During a 10 year trial by the World Health Organisation during the 1970s, >8,000 Chinese men took part in a trial of gossypol's contraceptive effects, taking 20 mg/day for up to 2 years. It was found to be an effective anti-spermatogenic agent, reducing the sperm count to below 4 million/cm^3 in 99.9% of patients.[2] However, the occurrence of irreversible infertility and hypokalaemia (fall in blood potassium levels), although rare, were considered unacceptable and gossypol trials were halted in 1986. Since then, gossypol has remained of interest, but its low aqueous solubility and relatively high toxicity at the dose required for anticancer activity are proving difficult to overcome. The (*R*)-(–)-enantiomer of gossypol exhibits the greater cytotoxicity.

Atropisiomers: assigning the stereochemistry of gossypol

How do we assign the stereochemistry of this compound? Actually, we follow rules similar to those used for alkenes to assign *R* or *S* as appropriate. Looking at the structure below on the left, Figure 3.21, we can see two sentinel groups (OH and CH_3) on one naphthyl group in the plane of the paper that can be prioritised and labelled as 1 and 2, respectively, while the two sentinel groups (OH and CH_3) on the other naphthyl group lying in front and behind the plane of the paper can be prioritised and labelled 3 and 4, respectively. These are viewed so that groups 1 and 2 are horizontal and groups 3 and 4 are vertical. As the priority 4 group is at the back, displayed as shown, it can be ignored; the direction of 1 to 2 to 3 is anticlockwise and shows that the enantiomer of gossypol shown on the left is the (*S*)-enantiomer.

[2] 4 Million sperm/ cm^3 may seem rather high, but when the sperm count is usually 20 – 150 million/ cm^3, this is considered infertile.

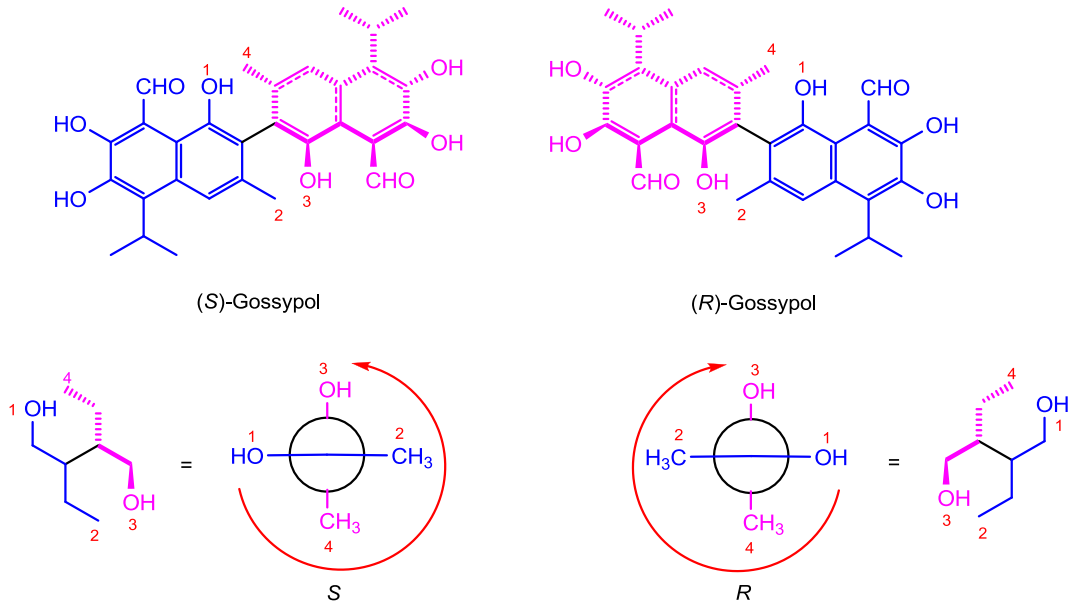


Figure 3.21 Nomenclature for atropisomers, using gossypol as an example.

Calculations involving the specific rotation

In chapter 3, we met the equation that relates specific rotation (of plane polarised light) to observed rotation, concentration and the length of the sample vessel through which the plane polarised light passes: equation 3.1.

$$[\alpha]_D = a / c.l \quad \text{at temperature T } (^oC) \qquad \textbf{Equation 3.1}$$

Where $[\alpha]_D$ = specific rotation in degrees (using the D line from a sodium light source at 589 nm)
a = observed rotation
c = concentration of sample solution (in g / cm^3)
l = path length in dm

Specific rotation calculations of pharmaceuticals when concentration is expressed as a %

Sometimes in pharmaceutical analysis, we consider solutions of drugs for which the concentration is known as a %. A solid may be dissolved in a solvent giving % w/v (weight by volume) or a liquid could be dissolved in a solvent giving % v/v (volume by volume). In this case, equation 3.2 is used.

$$[\alpha]_D = 100\ a / c.l \quad \text{at temperature T } (^oC) \qquad \textbf{Equation 3.2}$$

Where $[\alpha]_D$ = specific rotation in degrees (using the D line from a sodium light source at 589 nm)
a = observed rotation
c = concentration of sample solution (in % w/v or v/v)
l = path length in dm

Example 1: 7.5 g of (*S*)-naproxen were dissolved in chloroform to make 50 mL of solution. The optical rotation of this solution at 20°C was +9.9° with a cell length of 10 cm. Calculate the specific rotation of (*S*)-naproxen.

Firstly, we need to calculate the concentration of the (*S*)-naproxen in the chloroform solution as a %, which means the number of g of (*S*)-naproxen in 100 mL solvent. 7.5 g in 50 mL is equivalent to 15g in 100 mL, so the solution has a concentration of 15 % w/v. Now we can use equation 3.2:

$$[\alpha]_D = \frac{100\ x + 9.9}{15\ x\ 1} = \frac{+990}{15} = +66^o$$

This corresponds to the specific rotation of (*S*)-naproxen given in Box 3.2 on page 72.

Calculating the % optical purity of an unequal mixture of enantiomers

A variation of equation 3.1 can be used to calculate the % optical purity of a mixture, Equation 3.3:

$$\text{Optical purity \%} = 100\ [\alpha]_{mixture} / [\alpha]_{pure\ sample} \qquad \textbf{Equation 3.3}$$

Example 1: a solution of (*S*)-ibuprofen is slightly contaminated with (*R*)-ibuprofen and gives a specific rotation of +46.1°. We know that the specific rotation of (*S*)-ibuprofen is +57.6° and, using equation 2, we can calculate how much of each enantiomer is present in this solution.

Optical purity % = 100 x 46.1 / +57.6 = 80.0 %

This tells us that there is 80 % excess of the (*S*)-enantiomer over the (*R*)-form; this is called the **enantiomeric excess**. The remaining 20 % is therefore a mixture with no optical rotation (or it would change the value of 46.1° already measured), meaning that there is 10 % of (*S*) and 10 % of (*R*)-ibuprofen in this remaining 20 %. Overall then, there must be 90 % of (*S*)-ibuprofen in the mixture and 10 % of (*R*)-ibuprofen.

Example 2: calculate the % of (*S*)-naproxen sodium salt in an aqueous solution of observed optical rotation -10.45° ($[\alpha]_D = -11^o$).

If we add these numbers to equation 3.3, we get:

Optical purity % = 100 x (− 10.45) / (− 11) = 95.0 %

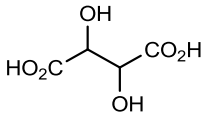
This tells us that there is a 95% enantiomeric excess of the (S)-enantiomer over the (*R*)-enantiomer and the remaining 5% must be made of an equal amount of both (*S*) and (*R*)-enantiomers. This means the sample contains 97.5% (*S*)-naproxen sodium salt and 2.5% (*R*)-naproxen sodium salt.

Specific rotation is altered by solvent

It is interesting to note that some chiral compounds display different rotation of plane polarised light is different solvents. For instance, we met ephedrine, a bronchodilator, in chapter 3 (in self-check 3.10 on page 76); it has a basic secondary amine group from which salts are readily formed. In self-check 3.10, you will have found that the active form of ephedrine is the (*R*,*S)*-enantiomer, also described as (−)-ephedrine due to its specific rotation of −34° (in aqueous solution). The optical rotation of the free base form of (*R*,*S*)-ephedrine (also used, very occasionally, as a bronchodilator) was originally measured on an ethanolic solution with a specific rotation of −5.5° at 22°C; later, the optical rotation was measured using an aqueous solution, leading to a specific rotation of +13.75° in water at 22°C.[3] Had the first ever optical rotation been made on an aqueous solution of the free base, ephedrine could have been recorded with the opposite sign! This makes the assignment seem rather 'hit and miss', but optical rotations were usually recorded on the hydrochloride salts of basic molecules and the $[\alpha]_D$ of (*R*,*S*)-ephedrine hydrochloride is negative in both aqueous and ethanolic solutions.

Activity

In the table below, you will see some selected data on tartaric acid; Louis Pasteur was working on the optically inactive sodium ammonium salt of tartaric acid in 1848 when he discovered that he could separate the (+) and (−) enantiomers due to visible differences in their crystalline forms. Using the principles discussed in the previous sections, consider the data in the table and identify a pair of enantiomers, a racemate, and an achiral compound. How can you explain the data provided for each molecule?



Tartaric acid

[3] This discovery was the result of some careful early chemistry: T.Q. Chou, *J. Biol. Chem*., 1926, **70**, 109 – 114 [www.erowid.org/archive/rhodium/chemistry/ephedrine.chou-jbc.html]. With improvements in equipment and more accurate measurements, the $[\alpha]_D$ of (−)-ephedrine (free base) in ethanol is now accepted as -6.3°.

Molecule(s)	$[\alpha]_D$ (25°C)	Melting point (°C)	Solubility (g/100g H_2O at 15°C)
(2R, 3R)-(+)-tartaric acid	+11.98°	170	139
(2S, 3S)-(−)-tartaric acid	-11.98°	170	139
(2R, 3S)-tartaric acid	0°	140	125
(±)-tartaric acid	0°	206	139

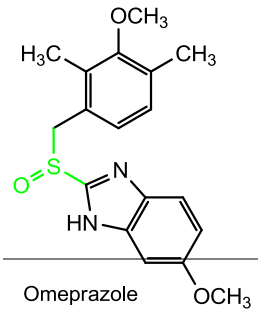
To look for a pair of enantiomers, we look for two molecules with mirror image stereochemical assignments with equal and opposite specific rotations. As they are enantiomers, they should have identical physical properties of melting point and solubility. Using these requirements, we can identify the pair of enantiomers as (2R, 3R)-(+)-tartaric acid and (2S, 3S)-(−)-tartaric acid.

A racemate is an equal mixture of enantiomers, so the optical rotation of one is cancelled out by together, leading to an optical rotation of 0. There are two entries in the table with a specific rotation of 0, so we will have to consider other factors. The melting point of a racemate is frequently higher than either of the separate enantiomers, due to the way the molecules pack together in the crystal lattice. This provides a clue that the last entry might be the racemate; the deciding evidence is the (±) prefix, as this means that there are both dextrorotatory and levorotatory molecules in the sample.

The meso-form is (2R, 3S)-tartaric acid. It has a specific rotation of zero, which is expected due to the internal symmetry in the molecule. The melting point and solubility are different to those of the separate enantiomers and the racemate.

Stereoisomerism based upon other atoms and systems

We have seen how stereoisomerism can arise from an asymmetrically substituted carbon atom. Are there any other ways in which stereoisomerism can arise? The answer is most definitely yes. Other substituted atoms that also adopt a stable tetrahedral shape, without spontaneous inversion, can exhibit similar stereoisomerism. Some examples follow.



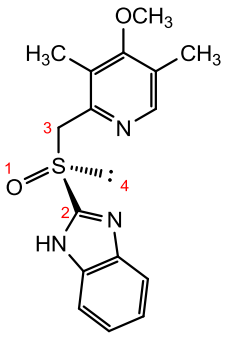
Omeprazole

Omeprazole is a proton pump inhibitor, so-called because this class of compounds blocks secretion of gastric acid in the stomach by inhibition of the enzyme hydrogen-potassium adenosine triphosphatase (H^+/K^+ ATPase) in the gastric parietal cells. It is used in the treatment of gastric and duodenal ulcers.

The key structural feature of omeprazole, in terms of both its stereochemistry and mode of action, is the sulfine group.

When you look at this structure, you can see three atoms attached to the S atom; they give the appearance of being planar, but the S atom has a tetrahedral shape. What occupies the 4th site?

The lone pair on the S atom occupies an orbital and gives rise to the tetrahedral shape of this atom and, hence, we have the requirements for chirality: an asymmetrically substituted tetrahedral atom. Now we need to draw it in 3D rather than the 2D representation above.



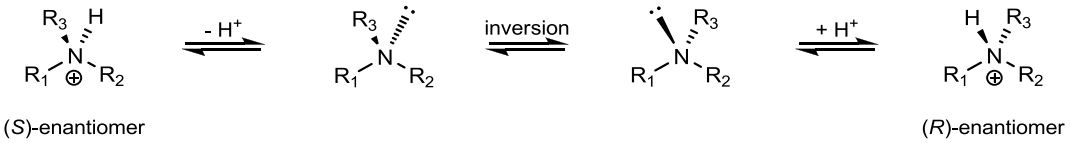
And, we can use the IUPAC nomenclature to assign the stereochemistry of such molecules! The lone pair has no atomic number ($Z = 0$), so it takes lowest priority.

The active enantiomer of omeprazole that reacts with, and inhibits, the H^+/K^+ ATPase enzyme is shown here. Use the same IUPAC rules to assign the stereochemistry and you will find that this is (*S*)-omeprazole (marketed as Esomeprazole®).

Answer: having identified the sulphur atom (S) as the chiral atom, assign the priority numbers: O 1, aromatic C 2 (has 3 bonds to N), CH_2 3, lone pair 4; the priority 4 lone pair is at the back so can be ignored. The sequence 1 to 2 to 3 goes anticlockwise, therefore this has *S* stereochemistry.

In this example, we have seen how omeprazole can exist as two possible stereoisomers due to the asymmetric substitution and tetrahedral shape of the sulfoxide group. This example also showed that a lone pair can form one part of a tetrahedron and contribute to the stereochemistry of an S atom. Sulfoxides in general exhibit chirality if not symmetrical.

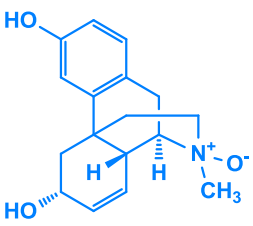
You may be wondering if the same applies to amines, with a trisubstituted N atom, which also bears a lone pair in the 4th position of a tetrahedron. The same theory applies, but amine chiral centres are usually able to interconvert rapidly, so amines are rarely chiral. Even protonated asymmetrically substituted tertiary amines very seldom permit isolation of the stereoisomers, as the acid-base equilibrium of a protonated amine allows for loss of a proton, inversion due to a low energy barrier, and then reprotonation, Scheme 3.4.



Scheme 3.4 The ready acid-base equilibrium of amines at approximately neutral pH facilitates inversion of chirality.

In this example, the H atom starts at the back of the structure; if the priority order is $R_1 > R_2 > R_3$, then this is the (*S*)-stereosiomer. After loss of the proton, inversion and reprotonation, the H atom is now at the front of the structure; although the order of priority groups looks anticlockwise, the lowest priority group is at the front, so we are viewing it from the wrong face. If we imagine viewing this molecule from the other face, so that the H is at the back, we find it has become the (*R*)-enantiomer and the chirality has inverted.

It would be logical to think that the enantiomers of asymmetric quaternary amines would be separable, as the fourth group attached to the chiral N atom is not H (so it cannot be lost to allow inversion); however, a phenomenon called tunnelling can occur, which allows the energy barrier to inversion to be by-passed and facilitates chiral inversion at N atoms. Tunnelling is only rarely seen in pharmaceuticals, so we will not consider it further here. Suffice to say that chiral quaternary amines are rare. One example of a chiral quaternary amine that is separable from its stereoisomers is provided by genomorphine.

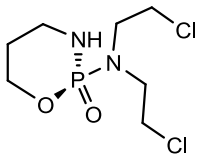


Genomorphine

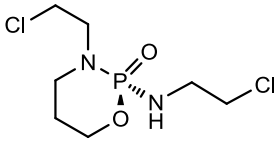
Although not used clinically, genomorphine (morphine-*N*-oxide) has potent anti-tussive properties (in other words, acts as a cough suppressant).

In this molecule, the quaternary N atom has four different atoms or groups attached to it and cannot invert, so its stereochemistry is fixed. There are 6 chiral centres, including the N atom, in genomorphine; can you identify them? Only one of the possible stereosiomers is shown here – the active form.

Many phosphorus (V) compounds have a tetrahedral arrangement of four substituents, from which chirality can arise; examples are cyclophosphamide and the related ifosfamide, Figure 3.22, which are used clinically as anticancer agents.



Cyclophosphamide



Ifosfamide

Figure 3.22 The chiral phosphate anticancer agents, cyclophosphamide and ifosfamide.

Only one enantiomer of each is drawn here, can you assign the chirality at each P atom? Interestingly, (*R*)-ifosfamide is linked with more rapid activation to the active agent and fewer adverse effects, while the (*S*)-enantiomer is activated more slowly and results in more metabolites linked to myelosuppression; however, both cyclophosphamide and ifosfamide are administered as the racemate (the 1:1 mixture of (R)- and (S)-enantiomers).

Helices

In chapter 3, we also met chiral helices, such as are found in proteins (the α-helix is an important form of secondary structure in proteins) and in double stranded DNA. How do we assign them as right or left handed? A right handed helix proceeds clockwise from the top, while the left handed helix proceeds anticlockwise from the top.

Did you know.....?

Spiral staircases in old castles are usually left-handed (you walk clockwise ***up*** them). It was easier for right-handed knights to defend a castle from above if the staircases were left-handed. A right-handed person going up the stairs has the centre column in the way of their sword-arm, putting them at a disadvantage.