Answers to end-of-chapter questions

1) Target specificity and selectivity refers to the ability of a drug to distinguish between different molecular targets, whether these targets be totally different in nature or slight variations of the same target. For example, drugs can show specificity or selectivity between different types and subtypes of receptor. The ability of a drug to distinguish between different targets is important since it results in more specific pharmacological and physiological effects, with fewer side effects (see also section 12.2).

2) The advantages of natural products as lead compounds are as follows:

- There is a greater chance of finding physiologically active compounds in nature since there is often an evolutionary advantage in an organism producing a physiologically active compound (e.g. as a defence chemical).
- Many natural compounds have totally novel structures which have not been synthesised before.
- Many natural compounds (e.g. toxins) are quite complex in nature with a highly rigid structure where the compound is locked into the active conformation or limited to a relative few number of conformations. This can result in high potency and selectivity for the target.

The disadvantages include the following:

- The natural product may only be present in small quantities in the natural source, restricting its availability.
- Isolating and purifying a natural product from its natural source tends to be slow, tedious and costly.
- The complexity of many natural products makes their synthesis impractical on a commerical basis. This in turn makes it difficult to produce analogues of the natural product.

See also sections 12.4.1 and 12.4.2.

3) There is a constant battle going on in the microbiological world between different microorganisms. Fungi and bacteria have to compete with each other for available nutrients and if one or the other gains some sort of advantage over the other, it could become dominant and a 'winner takes all' situation may arise. In general, fungi are slower growing than bacteria, which means that bacteria should be more likely to gain dominance. However, many fungi can counteract this disadvantage by producing antibacterial agents which either kill competing bacterial cells or slow down their growth. This allows fungal cells to compete with bacterial cells on a more even footing for available nutrients. There are significant differences between the cellular structures of fungi and bacteria. Fungal cells are eukaryotic and are more complex than the prokaryotic cells of bacteria, and so it is possible for fungi to produce agents which affect features of bacterial cells that are not present in fungal cells. Alternatively, antibacterial agents produced by fungal cells may disrupt a biochemical process that takes place in bacterial cells but not in fungal cells. Since fungal cells and mammalian cells are both eukaryotic in nature, there is a good chance that some of

these antibacterial agents may be used in medicine without serious side effects (see also section 12.4.1.2).

4) Many corals contain inflammatory agents which result in painful skin reactions if divers or snorkellers receive a coral cut. Reactions such as this indicate the presence of pharmacologically active compounds which could act as lead compounds for specific targets in the body.

5) Understanding how the target works would be the first stage in initiating a project. The endogenous ligand which normally activates the receptor could be identified. This then could be used as the lead compound for an antagonist which would bind to the receptor but not activate it. There are several examples of agonists which have been used as lead compounds for the design of antagonists (see section 12.4.5).
Another approach would be to study the reaction catalysed by the tyrosine kinase portion of the kinase receptor, and in particular the active site of the enzyme. A knowledge of how the protein substrate and ATP are bound to the active site would help in designing enzyme inhibitors which bind to the ATP binding region or the protein substrate binding site. This has been carried out successfully with several protein kinases with the help of X-ray crystallography and molecular modelling (see section 21.6.2).
Finally, one could carry out screening tests on different libraries of compounds produced by combinatorial and parallel synthesis to see if any of the compounds present show activity.

6) The genetically engineered form of the protein provides a water soluble preparation of the target enzyme which could be used for a direct enzyme assay. A standard phosphorylation reaction would be carried out on a protein substrate using the enzyme as catalyst, and where the level of substrate or product present could be measured. Potential inhibitors could then be introduced to see how effectively they inhibit this standard reaction. This test would detect effective enzyme inhibitors and allow medicinal chemists to identify a pharmacophore for those inhibitors. The results would not be complicated by other factors, such as the ability of agents to cross cell membranes.

The cell assay measures total tyrosine phosphorylation in the presence of epidermal growth factor. In this assay, the tyrosine kinase receptor is embedded in the cell membrane with the kinase active site located intracellularly. This assay would test the ability of an inhibitor to cross the cell membrane in order to reach the target active site, as well as its ability to inhibit the phosphorylation reaction. If this assay was not carried out, a great deal of time and effort could be spent optimising a highly active inhibitor which could not be used clinically since it cannot reach its target.

The *in vivo* study on mice measures whether a test compound reaches the tumour and can stop the growth of the tumour by inhibiting the kinase active site. This establishes whether the test compounds are stable to drug metabolism, whether they have a reasonable lifetime or are excreted too quickly, whether they actually reach the tumour, and whether inhibition of the target has the desired physiological effect.