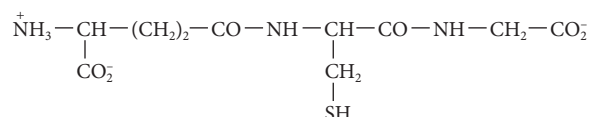


Solutions to problems for Chapter 1

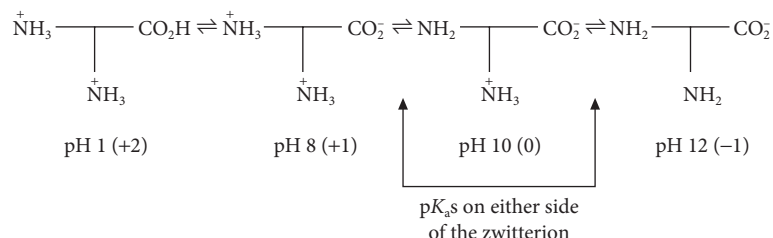
1.1 Ornithine should be similar to Lys: a polar, charged, amino acid. The pK_a of the side chain amino group should be about 10.5. The amino group can act as a nucleophile and a hydrogen bond donor.

1.3 Glutathione is unusual in that the linkage between the Glu and the Cys is a peptide bond formed between the γ -carboxyl of Glu and the α -amino group of Cys. Under oxidizing conditions, a disulphide bond can be formed between the Cys side chains of two glutathione molecules ($2\text{GSH} \rightleftharpoons \text{GSSG} + 2\text{H}$). The GSH/GSSG system is an important intracellular redox system. The structure of glutathione is shown below.



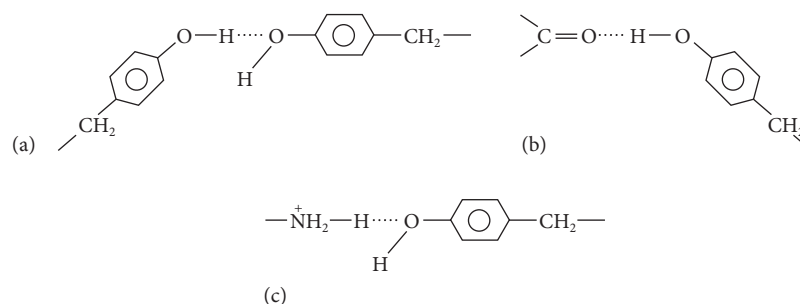
1.5 Side chains which would be expected to coordinate with Zn^{2+} include those of Cys and Met (the latter would be less likely because of methyl substitution on the S atom which would increase its bulk), and the side chain of His, which could provide an uncharged N atom. The side chains of Lys and Arg would be positively charged and unlikely to coordinate with Zn^{2+} , but it is possible that the uncharged N atoms of the amide side chains of Asn and Gln could do so. The so-called 'zinc finger' motif of some DNA-binding proteins involves coordination of Zn^{2+} by two Cys and two His side chains.

1.7 The predominant forms of Lys at different pH values carry the charges pH 1 (+2), pH 8 (+1), pH 10 (0), and pH 12 (-1). The pI is the average of the two pK_a s around the zwitterionic form (i.e. $(9.1 + 10.5)/2 = 9.8$). At this pH, there is a negligible contribution from the (+2) form, so it makes no difference to the overall charge on the amino acid. Representations of the predominant forms are shown below.



1.9 Consider the equilibrium $\text{Lys-NH}_3^+ \rightleftharpoons \text{Lys-NH}_2 + \text{H}^+$. A neighbouring Arg (positive charge) will shift the equilibrium to the right, since it will be unfavourable to have two positive charges close. This will lower the pK_a , i.e. make dissociation more favourable. A neighbouring Asp (negative charge) will shift the equilibrium to the left, since it will be favourable to have opposite charges close. This will raise the pK_a , i.e. make dissociation less favourable.

- 1.11** Using the relationship $-\Delta G^0 = RT \ln K_{eq}$ and a value of $\Delta G^0 = +20 \text{ kJ mol}^{-1}$ for the *trans* \rightleftharpoons *cis* equilibrium, K_{eq} is calculated to be 4.25×10^{-4} . Thus approximately 0.04% of peptide bonds should be in the *cis* form.
- 1.13** The molar extinction coefficient at 280 nm = $(5 \times 5690) + (14 \times 1280)$ = $46370 \text{ M}^{-1} \text{ cm}^{-1}$. Thus the A_{280} of a 1 mg mL^{-1} solution in a cuvette of 1-cm pathlength is $46370/36712 = 1.263$. The A_{280} of a 0.32 mg mL^{-1} solution = $1.263 \times 0.32 = 0.404$.
- 1.15** The helical wheel projections of a type similar to that shown in Fig. 1.14 show that in sequence (a) both faces of the helix are mainly non-polar, so the helix is likely to be buried in the interior of the protein, in sequence (b) one face of the helix is polar and the other non-polar, so the helix is likely to be at a polar/non-polar interface, and in sequence (c) both faces of the helix are polar, so the helix is likely to be completely exposed to the solvent.
- 1.17** The arrangements are (a) one Tyr as H-donor, the other as acceptor, (b) Tyr as H-donor, C=O as acceptor, and (c) Tyr as acceptor, $-\text{NH}_3^+$ as donor. The structures of these are shown below.



- 1.19** Ionic interactions are strongest when the ionic strength (I) of the medium is low, and become progressively weaker at higher I due to competition from the ions of the medium. Hydrophobic interactions are stronger at high I (the non-polar side chains are excluded to an even greater extent) and become weaker as I is decreased. Thus in ion-exchange chromatography, adsorption is carried out at low I and desorption is brought about by increasing I . In hydrophobic interaction chromatography, adsorption is carried out at high I and desorption by decreasing I . It may be necessary to decrease the polarity of the solvent by adding an organic solvent such as ethylene glycol (1,2-ethanediol) in order to achieve more complete desorption.