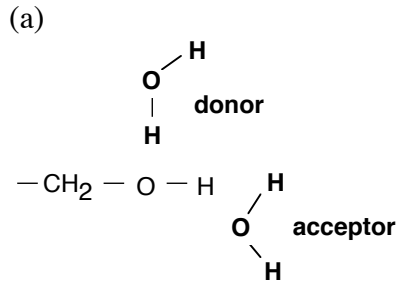


Biochemistry – Problem set 1

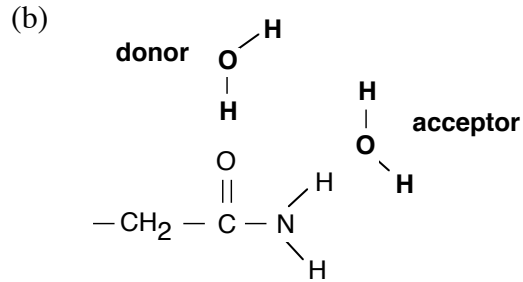
**Problem Set 1 - Solutions**

1. (8 points, 10 min) Identify the following amino acid side chains (1 pt each) and draw one hydrogen bond likely to form between each side chain and water (1 pt each): (1 is sufficient)

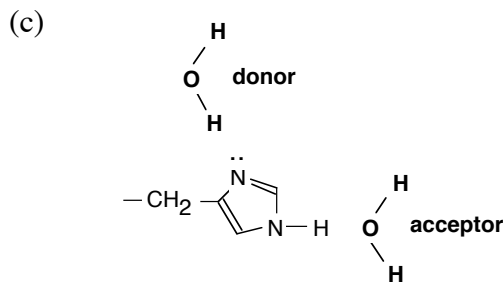
[Draw the waters as “H-O-H” and indicate for each whether the water is the H-bond donor or acceptor].



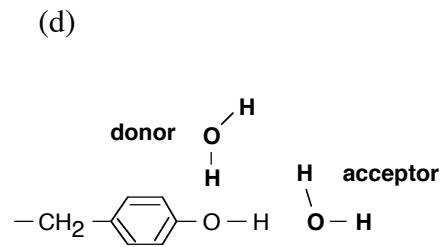
**Serine**



**Asparagine**

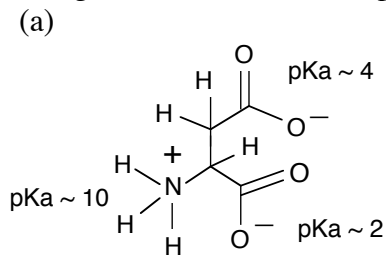


**Histidine**

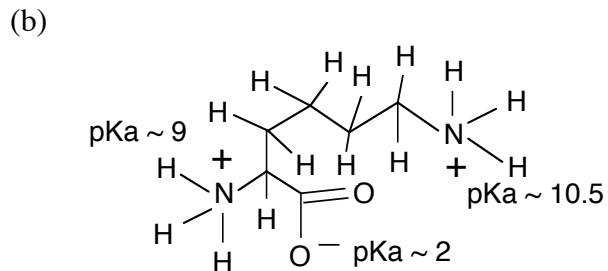


**Tyrosine**

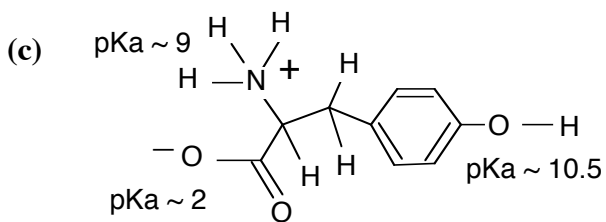
2. (15 points, 15 min) The side chains of several amino acids possess ionizable side chains. Identify the following amino acids (1 pt each), add any protons that may be missing from each structure (1 pt), and draw each in its correct ionic form at pH 7.0 (see table 3.2 in Campbell for  $K_a$  values). Label *all* ionizable groups with their  $pK_a$  values (1 pt), include electrostatic charges (1 pt) and indicate the net charge of the amino acid (1 pt).



**Aspartate (-1)**

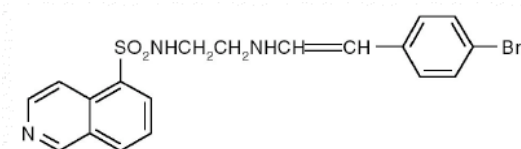


**Lysine (+1)**



### Tyrosine (0)

3. (4 points, 5 min)
- (a) **Water** is a better solvent than methanol because **its dielectric constant is higher; hence the attractive force between the charges on  $\text{Na}^+$  and  $\text{Cl}^-$  are weaker in water** than in methanol. (2 pts)
- (b) The drug is more likely to be soluble in **ethanol** than water because **it is hydrophobic**. (2 pts)



4. (9 points, 25 min) You are required to make 100 ml of a 0.05 M potassium phosphate solution for a biological experiment at pH 7.4. You only have the fully protonated form of this acid ( $\text{H}_3\text{PO}_4$ ) in the lab, plus 1 M solutions of HCl, acetic acid, NaOH and KOH.
- (a) Phosphate is a good choice **because one of its  $\text{pK}_a$  values is within one unit of the desired pH**.
- (b) You would need **0.005 mole** of phosphate (0.1 L x 0.05 mole/L).
- (c) Since you are beginning with the acidic form, **base** will have to be added to raise the pH. **KOH is the best choice since the buffer is to be potassium phosphate** rather than *sodium* phosphate.
- (d) What volume of the base would be required to adjust the pH of the phosphoric acid solution to 7.4?

This problem is somewhat tricky because it involves a triprotic acid and the desired pH is above the first  $\text{pK}_a$ . Therefore it is necessary to fully titrate the first group and then partially titrate the second. The calculation can be approached by considering the acid to be monoprotic and then accounting for the first titration.

The known quantities are:  $\text{pK}_a = 7.2$ ;  $[\text{A}_T] = 0.05 \text{ M}$ ; desired pH = 7.4.

$$\text{First calculate R: } R = 10^{[\text{pH} - \text{pK}_a]} = 10^{(7.4 - 7.2)} = 10^{0.2} = 1.6$$

Then calculate  $[\text{HA}]$  and  $[\text{A}^-]$

$$[\text{HA}] = [\text{A}_T] \times 1 / (1 + R) = (0.05 \text{ M}) \times 1 / 2.6 = 0.02 \text{ mole/L}$$

$$[\text{A}^-] = [\text{A}_T] \times R / (1 + R) = (0.05 \text{ M}) \times 1.6 / 2.6 = 0.03 \text{ mole/L}$$

Since we have a total of 100 mls, we only need 1/10 of each of these. In moles:

$$\text{moles HA} = 0.002$$

$$\text{moles A}^- = 0.003$$

To convert the correct amount of HA to  $\text{A}^-$  it is necessary to add 0.003 mole of KOH. **However**, since this is a diprotic acid, we have to fully deprotonate the first acidic group ( $\text{pK}_a \sim 2$ ) to reach the desired pH. Therefore it is necessary to add 0.008

mole (0.005 mole to fully deprotonate the first group + 0.003 mole) of KOH, or **8 mls** of the 1 M KOH solution. As the base is added, the acid will be deprotonated as follows:



until the correct ratio of  $\text{HPO}_4^{2-} / \text{H}_2\text{PO}_4^-$  is attained.

5. (10 points, 25 min) A polypeptide contains *two* Arginine residues and *one* Histidine residue, and no other amino acids with ionizable sidechains. The  $\text{pK}_a$  of the sidechains of the Lysine residues are 12.5 and that for the Histidine is 5.0. Assume that the  $\text{pK}_a$  of the mainchain carboxylic acid group is 2.0 and that of the amino group is 10.0.

(a) *Sketch* a titration curve for this protein.

There are *five* ionizable groups, the carboxy-terminus ( $\text{pK}_a=2.0$ ), the His sidechain ( $\text{pK}_a=5.0$ ), the amino terminus ( $\text{pK}_a=10.0$ ), two Arg sidechains ( $\text{pK}_a=12.0$ ), and four different  $\text{pK}_a$  values. Therefore, there should be four inflection points and the complete titration will require five equivalents since five protons have to be removed from this polypeptide. Note that the  $\text{pH} = \text{pK}_a$  when 0.5 (carboxy terminus), 1.5 (His), 2.5 (amino terminus), and 4 (two Arg residues) equivalents have been added.

(b) Calculate the fraction of the Histidine side chain that is protonated at pH 6.0. (3 pts)

Use the same approach as with the buffer calculation. Obtain R first, and then calculate [HA].  $R = 10^{6.0-5.0} = 10^1 = 10$ , therefore  $f_{\text{HA}} = 1/(1+R) = 1/11 = 0.09$ . Therefore **9% of the sidechain is in the protonated form**, as expected, since the pH is one unit above the  $\text{pK}_a$ .

(c) What is the normal  $\text{pK}_a$  for a Histidine sidechain? Suggest a reason why it is lower than normal in this particular polypeptide. (2 pts)

**The normal  $\text{pK}_a$  for His is 6.0.** Since the His is a stronger acid in the polypeptide than in solution, its deprotonated form must be stabilized, **likely due to a favorable electrostatic interaction with the two Arg residues.**

