بسم الله الرحمن الرحيم

BioChemistry | Lecture 4

pH and Buffers pt.2

Written by : Abdallah Al-Abdallat
Rayan Theeb

Reviewed by : Ibrahim Al-Zoubi

Henderson-Hasselbalch Equation

$$HA \longleftrightarrow H^+ + A^-$$

$$K_a = \frac{[H^+][A^-]}{[HA]}$$

$$[H^+] = \frac{K_a[HA]}{[A-]}$$

$$\log[H^+] = \log K_a + \log \frac{[HA]}{[A-]}$$

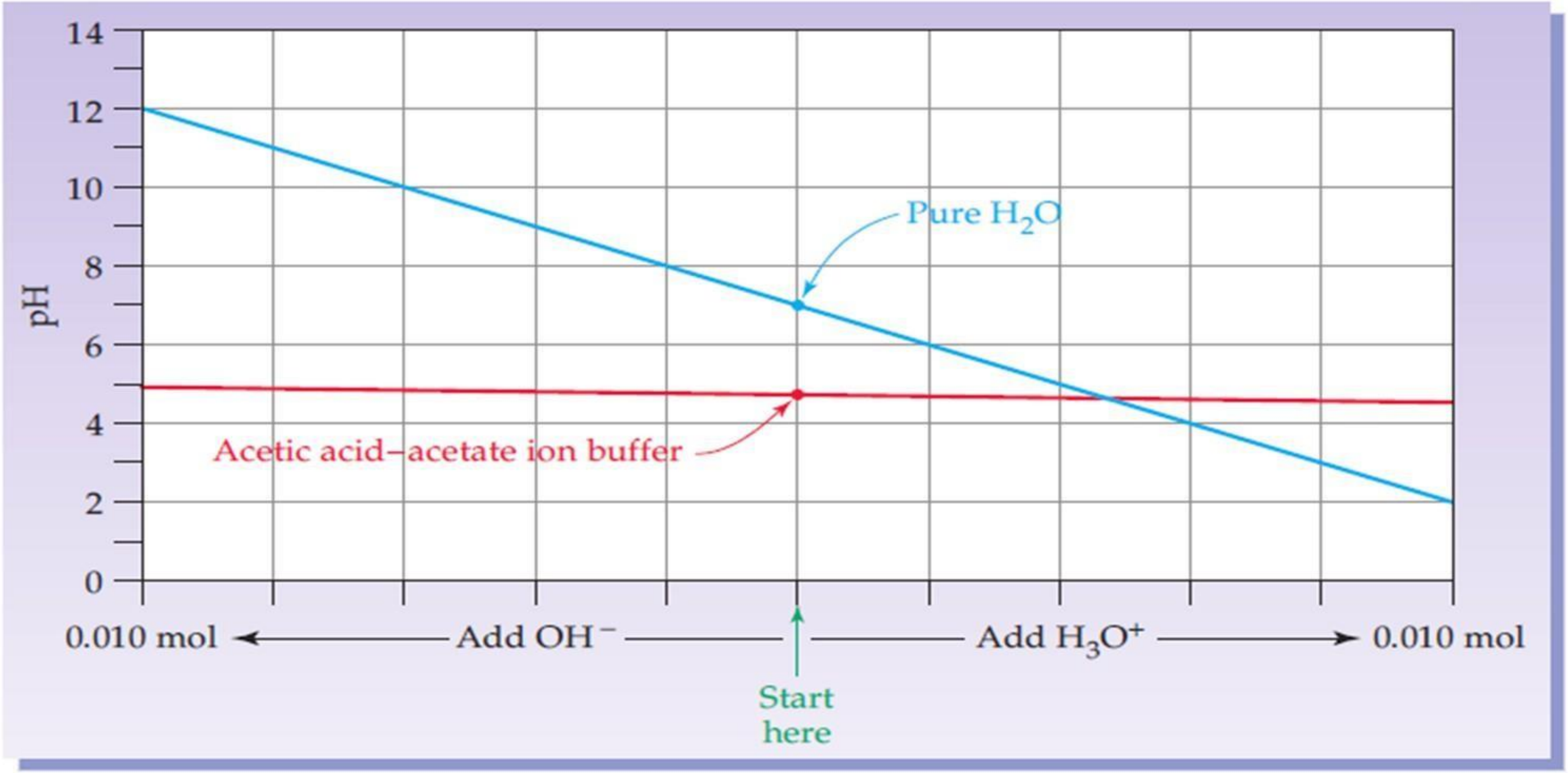
The purpose of this equation is to relate pH, pKa, and the ratio of the conjugate base to the acid. Since pKa is constant for a given acid, the equation shows that pH depends directly on the ratio of the conjugate base [A−] to the acid [HA]. Therefore, changing the pH affects this ratio, and vice versa.

$$pH = pK_a - \log \frac{[HA]}{[A]}$$

or

$$\boxed{pH = pK_a + \log \frac{[A^-]}{[HA]}}$$

A comparison of the change in pH (water vs. acetic acid)



0.010 mol of base are added to 1.0 L of pure water and to 1.0 L of a 0.10 M acetic acid 0.10 M acetate ion buffer, the pH of the water varies between 12 and 2, while the pH of the buffer varies only between 4.85 and 4.68.

- The previous chart relates the changes in pH(y-axis) when you add a strong acid/strong base as it appears on the x-axis.

- When you add a strong acid or base to the pure water, the pH will change drastically with absolutely no resistance.
- On the other hand, if we add H^+ or OH^- to acetic acid-acetate ion buffer, there will be only a very slight change.
- This is because of the reversible reaction and the production of H^+.

- For the buffer to be effective, it requires relatively equal concentrations of the acid and its conjugate base.

What is a buffer?

Buffers are solutions that resist changes in pH by changing reaction equilibrium

They are composed of mixtures of a weak acid and a roughly equal concentration of its conjugate base

Weak Acid	Conjugate Base
CH_3COOH	CH_3COONa ($NaCH_3COO$)
H_3PO_4	NaH_2PO_4
H_2PO_4- (or NaH_2PO_4)	Na_2HPO_4
H_2CO_3	$NaHCO_3$

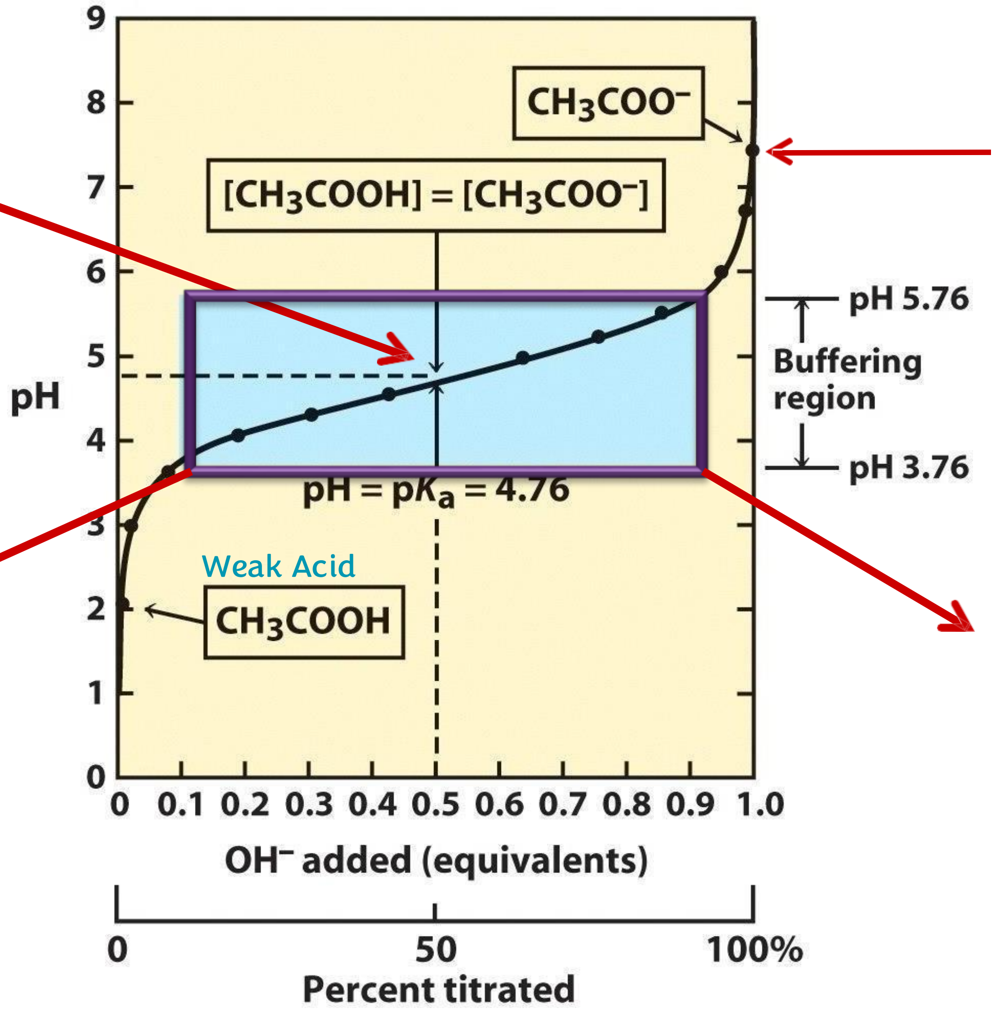
How do buffers work?

- Let's take a case where you add H+ (strong acid), it will bind with [A−], shifting the equilibrium towards making more of the [HA], so the extra H+ that you've added won't be effective and will result only in a very slight change in the pH.
- The other case is when we add OH− (a strong base), which binds with H+ to produce H2O, thereby decreasing the amount of H+. This results in a shift of the equilibrium towards the production of H+, thereby raising the concentration of [A−].

$$HA \longleftrightarrow H^{+} + A^{-}$$

Titration curve of buffer

What is the midpoint?

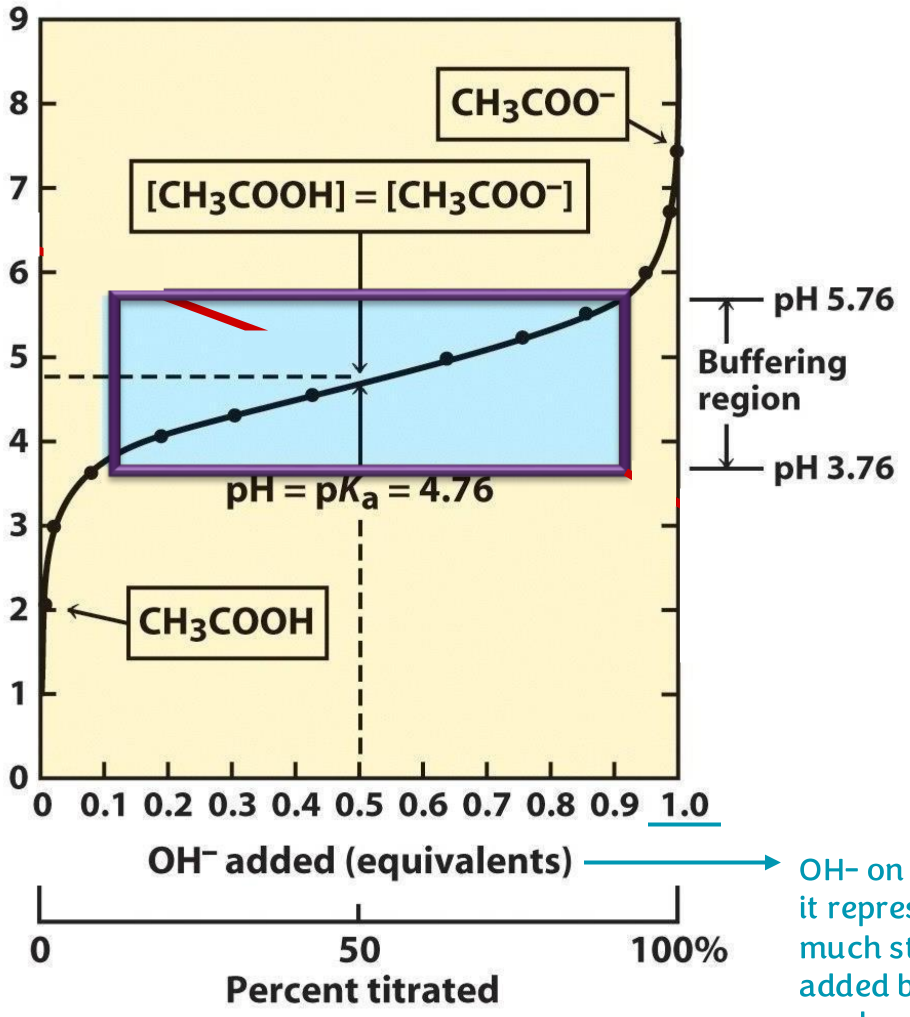


Equivalence point

Buffering capacity

What is the ratio of the conjugate base:acid at the different points?

Titration curve of buffer



pH on the y-axis

OH- on the x-axis, it represents how much strong base is added bit by bit to a weak acid

- The process of gradually adding a strong base to a weak acid solution to neutralize it is called titration
- The equivalents that are shown on the x-axis end with the number "1.0" as pointed, which means 1 full equivalent of base has been added, and it reached the equivalence point, where all the weak acid has been neutralized

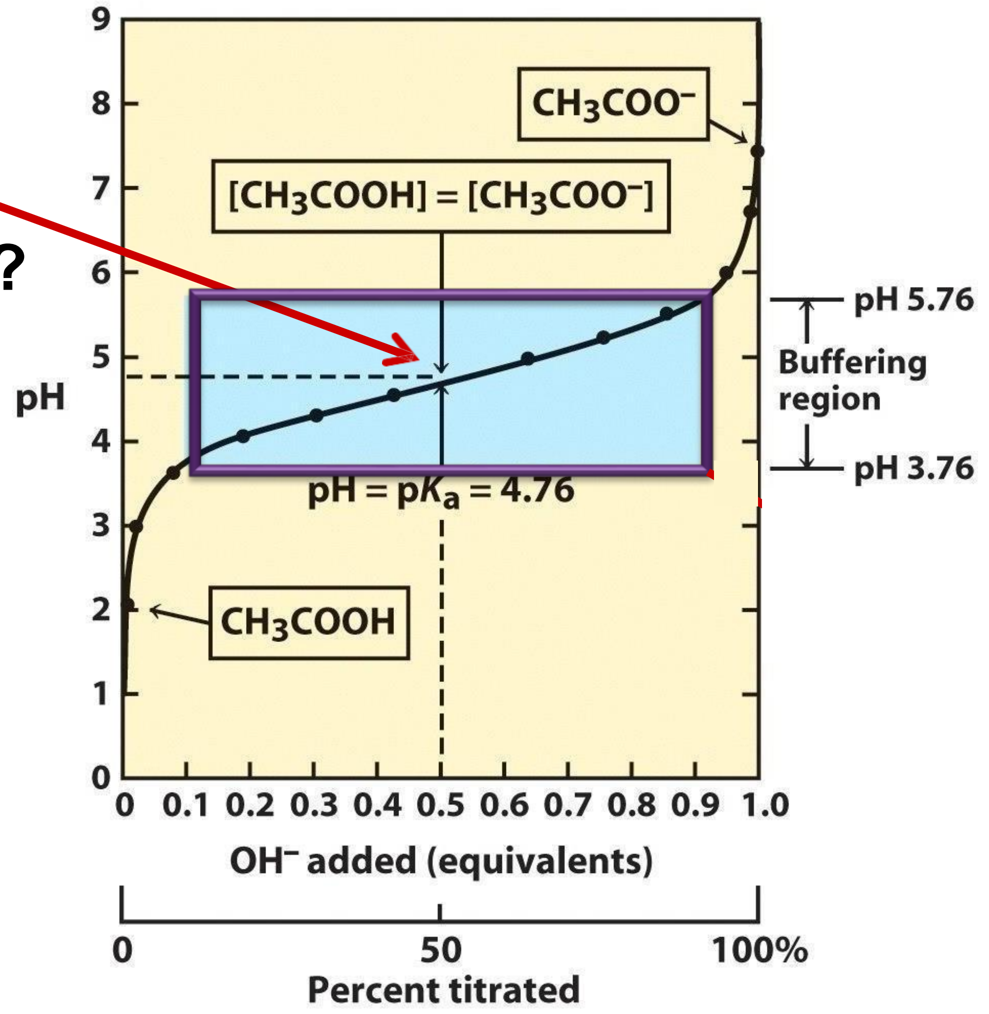
  More specifically, the number "1.0" shows that the acid is **monoprotic** (which means that it can donate 1 proton H+), if it went up to 2.0 it would mean a diprotic acid, and so on…

Titration curve of buffer

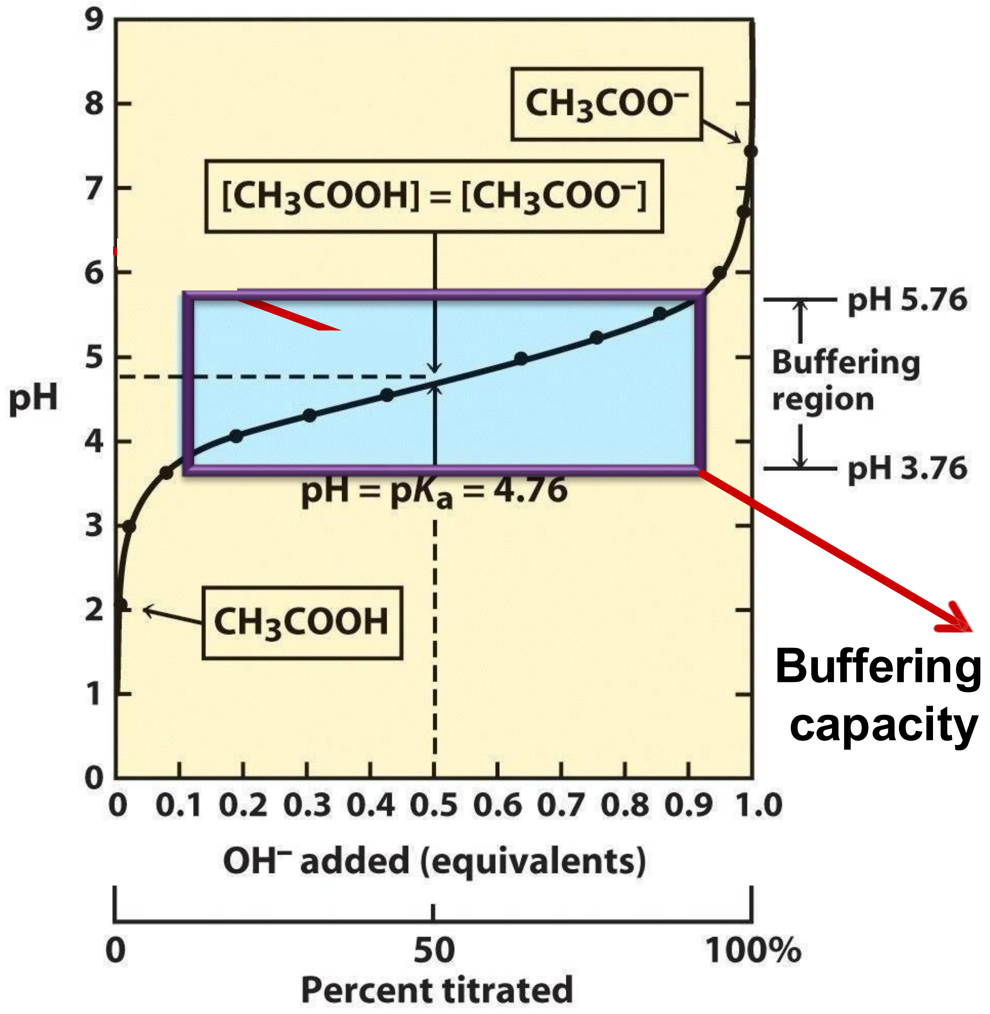
- It is where half the acid is neutralized, which means that 0.5 equivalents is added to the acid
- At this point 50% of the acid is still protonated (still in the acid form) and 50% is deprotonated (converted to the conjugate base)
- The ratio of the acid to base here is 1:1, thus pH=pKa
- So, what is the pKa? It is **the pH value** at which 50% of the weak acid is dissociated. This point is reached as you gradually add base to the solution, leading up to neutralization.

- While you are adding OH^-, it reacts with the free H^+ ions in the solution. This reduces the concentration of H^+, which disturbs the equilibrium. As a result, more of the undissociated acid (HA) dissociates to replace the lost H^+ ions, shifting the equilibrium toward more dissociation.

What is the midpoint?



Titration curve of buffer



- Buffers have a limited capacity, known as buffer capacity. This refers to the ability of the buffer to resist pH changes when small amounts of acid or base are added.

  For example, when acid (H^+) is added, it binds with the conjugate base (A^-) in the buffer, forming more of the weak acid (HA). This shifts the equilibrium to the left, helping to prevent a sharp increase in H^+, so the pH remains relatively stable.

  On the other hand, when a base (OH^-) is added, it reacts with H^+ ions to form water, As H^+ is removed, the weak acid (HA) dissociates more to replace the lost H^+, again helping to keep the pH stable.

- However, the buffer system can only resist pH changes effectively within its capacity. Once the concentration of the acid or conjugate base becomes too low, the buffer can no longer neutralize the added H^+ or OH^- effectively. At this point, the buffer is no longer effective, and pH will change significantly.

- So, for a buffer to work properly, it needs to be present in sufficient concentration, and it functions best within about ±1 pH unit of its pKa.

* For example:
A Buffer whose pKa is 4
And if pH is 3
Log([A-]/[HA]) will be -1
Which means that the ratio is 1:10
91% is acid
9% is base

* After this ratio(+-1) pH, the buffer can’t function effectively because we don’t have enough molecules to neutralise HA or A-

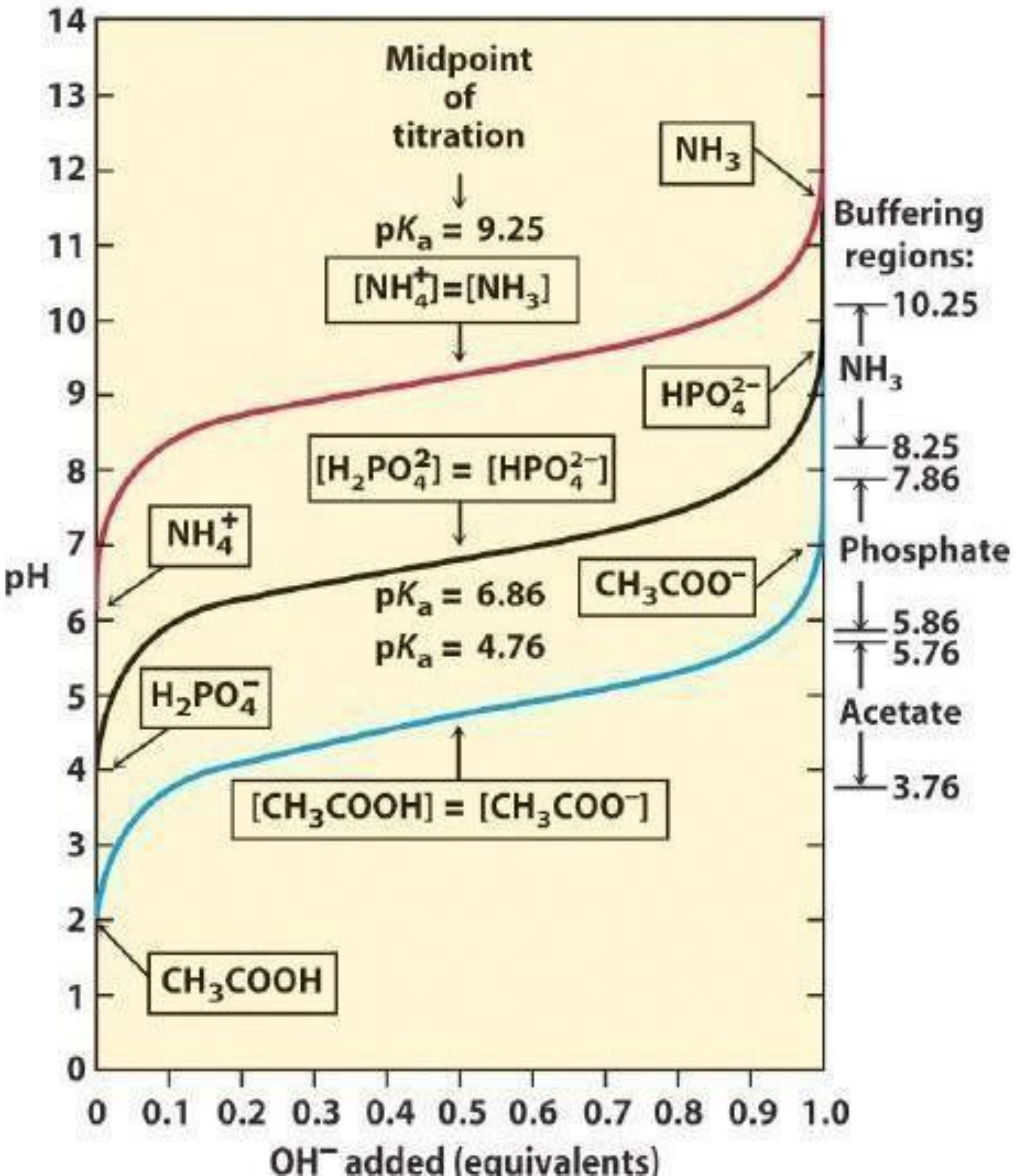
How do we make/choose a buffer?

A buffer is made by combining weak acid/base and its salt.

The ability of a buffer to function depends on:

- Buffer concentration To be in good values
- Buffering capacity (pKa of the buffer and the desired pH)

Choosing buffer depends on the pH of the environment where proteins are functional



Exercises

A solution of 0.1 M acetic acid and 0.2 M acetate ion. The pKa of acetic acid is 4.8. Hence, what is the pH of the solution? Final answer: 5.1

Without calculator:
pH = pKa + log([Conjugate base]/[acid])

$0<\log(0.2/0.1)<1 \ldots.\ 4.8<pH<5.8$

Predict then calculate the pH of a buffer containing

- 0.1M HF and 0.12M NaF? ($Ka = 3.5 \times 10^{-4}$) Ka = ([H+]*[F−])/[HF] Final answer: 3.54
- 0.1M HF and 0.1M NaF, when 0.02M HCl is added to the solution?

Ka = ([H+]*([F−]-[HCl])) /[HF]+[HCl]
Ka = ([H+]*0.08 / 0.12)

Final answer: 3.28

❖ The addition of 0.02 HCl will result in the presence of 0.02M of H+, which will react with F again, forming extra 0.02 HF and 0.02 missing NaF.

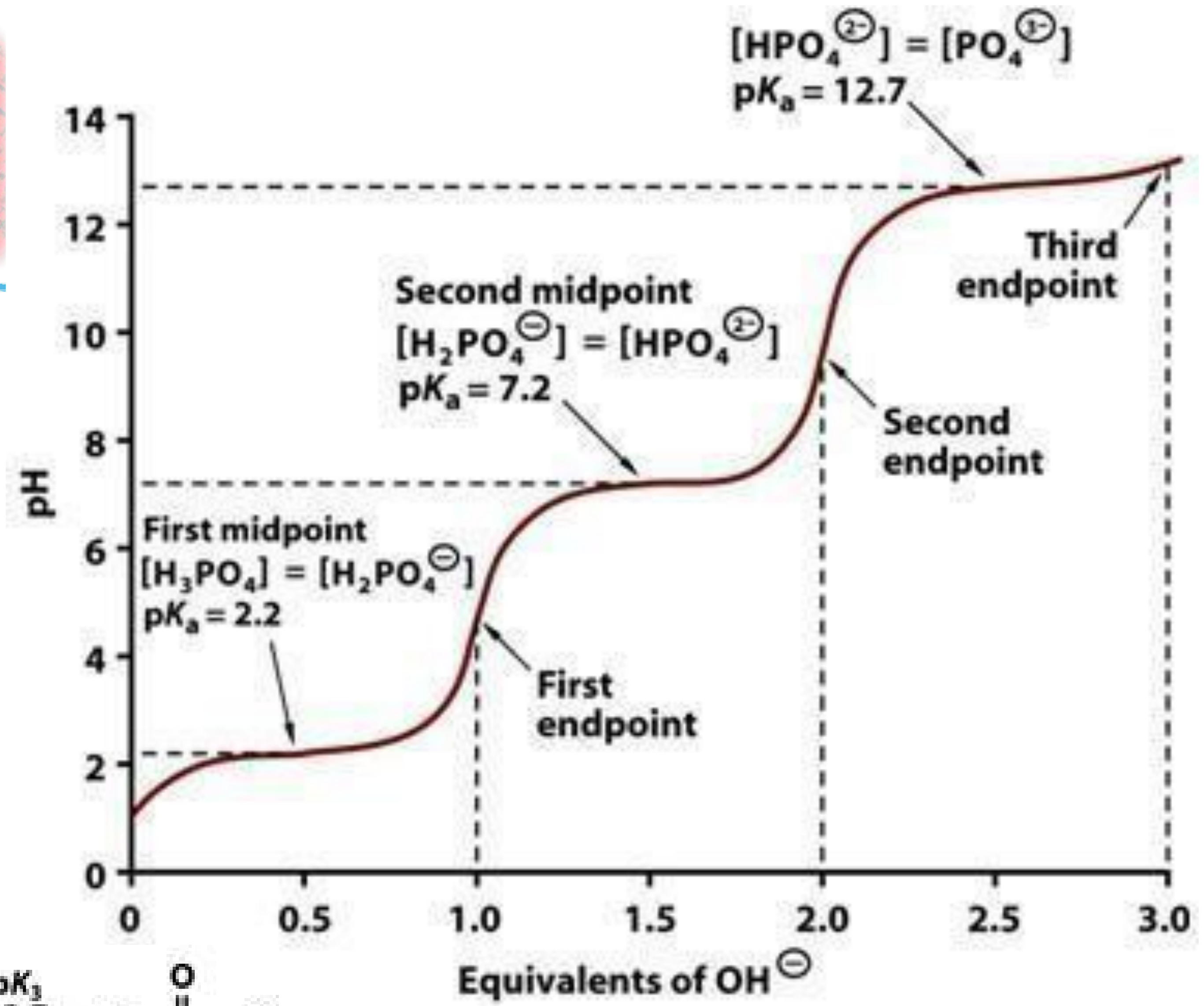
What is the pH of a lactate buffer that contain 75% lactic acid and 25% lactate? (pKa = 3.86)

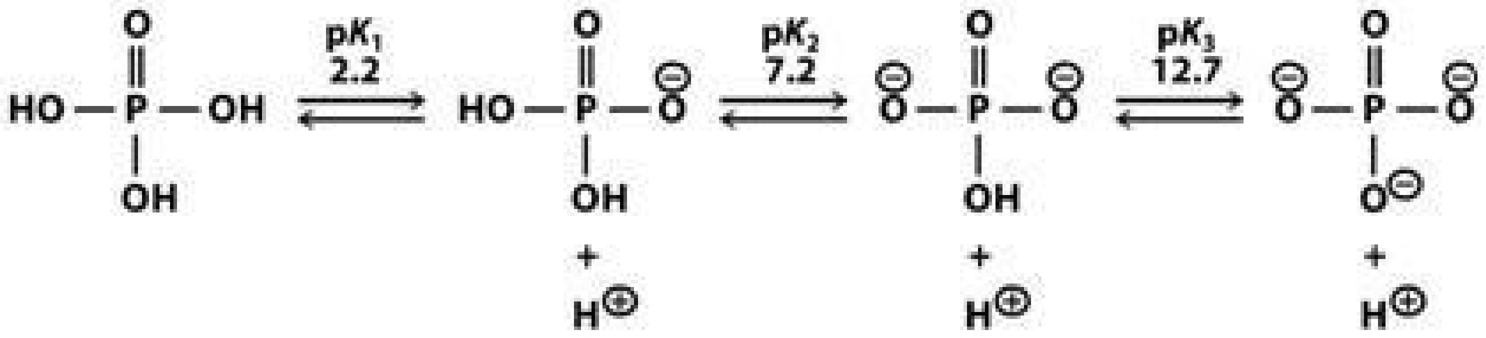
pH = pKa + log(25/75)
Log is negative….pH<3.86

Final answer: 3.38

Titration curve of phosphate buffer

Triprotic buffer
Release protons depending on pH.
If solution has many H+ , protons won't dissociate.
When H+ decrease, one of the protons will dissociate depending on the strength of bond which is represented by ka.
It has 3 pH ranges, so it covers most of pH values.
It is presented inside cells and Interstitial fluid





Exercises

A solution was prepared by dissolving 0.02 moles of acetic acid (HOAc; pKa = 4.8) in water to give 1 liter of solution. What is the pH? Final answer: 3.25

M(HOAc) = 0.02/1 = 0.02M

At equilibrium:

[H+] = [conj.base] = x

New M(HOAc) = 0.02 – x

Convert pKa to Ka

Calculate x by this equation

Ka = [Conj.base]*[H+] / [acid] … [conj.base] = [H+]

We assume x << 0.02

$Ka = x^2 / 0.02$

If $(x/0.02) < 0.05$

We neglect x

Otherwise, we recalculate x by this equation

$Ka = x^2 / (0.02-x)$

To this solution was then added 0.008 moles of concentrated sodium hydroxide (NaOH). What is the new pH? (In this problem, you may ignore changes in volume due to the addition of NaOH). Final answer: 4.62

[Acid] = (0.02-0.008)/1

[Conj.base] = (0.08)/1

Then, by Henderson-Hasselbalch equation

Buffers in human body (biological buffers)

Blood / Interstitial fluid / Intracellular fluid

- Carbonic acid-bicarbonate system (blood)

Around 7.4

- Dihydrogen phosphate-monohydrogen phosphate system (intracellular)

And Interstitial fluid

- ATP, glucose-6-phosphate, bisphsphoglycerate (RBC)

High concentration in body
They have negatively charged phosphate, so they bind H+

Any molecule that can bind to H+ can be a good buffer

- Proteins (extra- and intracellular) (why?)

In between amino acids, no ionizable groups, but on sides of proteins, there is carboxylic and amino groups which can bind to H+

Hemoglobin in blood →

Can bind H+ to specific histidine residues
Also, can bind to CO2 (acid, part of buffering system of blood) and form carbamate ion

Other proteins in blood and cells

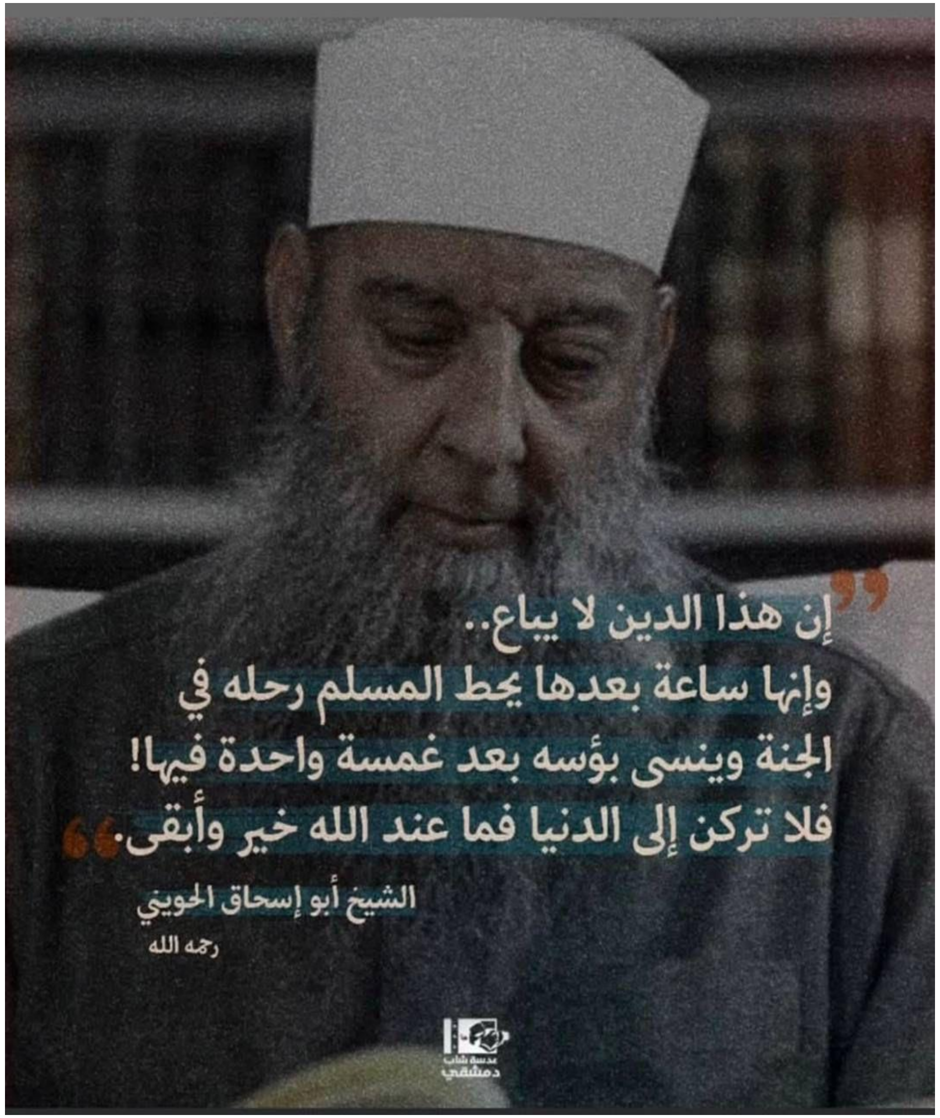
For any feedback, scan the code or click on it.

Corrections from previous versions:

Versions	Slide # and Place of Error	Before Correction	After Correction
V0 → V1	Slide 13 Question 2 ([HF] + [HCl])	0.012	0.12
V1 → V2			

Additional Resources:

رسالة من الفريق العلمي:



لا ترْجُ الرّاحة في دارٍ لم تخلق للراحة!
سُئل الإمامُ أحمد: متى الرّاحة يا إمام؟ فقال: عند أوَّلِ قدم تضعها في الجنَّة!
يا صاحبي: لا ترجُ الرّاحة في دارٍ لم تخلق للراحة!
ألفَ سنةٍ إلَّا خمسين عاماً كُذِّبَ نوح عليه السّلام!
وأُلقيَ خليل الله إبراهيم عليه السَّلام في النَّار!
وفي بطن الحوت لبث يونس عليه السَّلام!
بالمنشار نُشِرَ زكريا عليه السَّلام، ولبغيٍّ قُدِّمَ مهراً رأس يحيى عليه السَّلام!
رُجم نبيك ﷺ في الطائف، وحوصر في الشعب، وسال دمه يوم أحد!
هؤلاء هم صفوة الخلق، وتريد أن ترتاح أنت هنا!
سأل رجلٌ أحد التَّابعين: متى أطمئن؟ فقال له: إذا رأيتَ الصِّراط خلفك!

[من كتاب رسائل من التابعين]