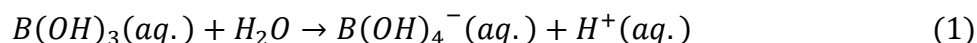


Borate buffer System Background

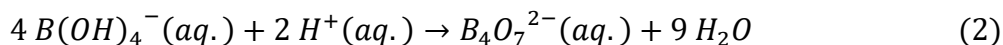
There are various procedures for making a borate buffer. One method is to prepare a working buffer by using H_3BO_3 and NaOH , or by using sodium tetraborate and HCl . Perhaps the most elegant method is to use sodium tetraborate also known as borax ($\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$) and H_3BO_3 . The boric acid-borate system is quite complex. We use this rather than other high pH buffers such as Tris or ammonium ion buffers because any free amines will act as quenchers of uranine. We cannot very well measure the quenching by iodide if the buffer we choose also acts as a quencher.

One fact that makes borate buffer particularly difficult is that orthoboric acid (H_3BO_3 , also written $\text{B}(\text{OH})_3$) and borax ($\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$) do not appear to be stoichiometrically related. However, if we treat a solution of boric acid with NaOH and then evaporate to dryness, we obtain borax ($\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$).

Boric acid actually ionizes by accepting a hydroxide ion (and thus an electron pair) from water, rather than by donating H^+ . This makes it a Lewis Acid:

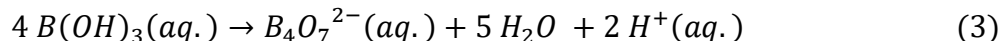


When protonated, the tetrahydroxyborate ion yields the tetraborate conjugate base



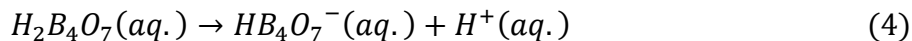
We see that boric acid is a Lewis Acid, not a classic proton donor, so these equations don't give us the typical Arrhenius acid-base equilibrium expressions. Notice here the tetraborate ion, $\text{B}_4\text{O}_7^{2-}$, which occurs also with the ionization of borax. In a borax-boric acid buffer solution (~pH 9), tetraborate and monohydrogen tetraborate are actually the primary species, as long as the boron concentration is greater than about 0.025 M.

In theory, it takes four moles of boric acid to yield one mole of $\text{B}_4\text{O}_7^{2-}$ in solution. If we quadruple the coefficients in Equation 1 to correspond with Equation 2, we get $4\text{B}(\text{OH})_3$. When we combine the two equations we obtain:

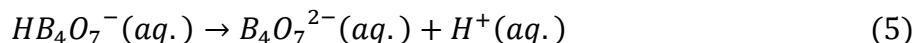


Notice that the H^+ is now on the right, which is consistent with the fact that boric acid yields an acidic solution. Because $\text{H}_2\text{B}_4\text{O}_7$ is not a strong acid, though, it does not completely ionize.

First ionization:



Second ionization:



These two equilibria look like what we would expect for a typical acid-base equilibrium, the so-called Arrhenius-Ostwald model. The second ionization (Equation 5) is the more important one for the buffer we will make since the monohydrogen tetraborate ion has a pK_a of 9. Although this is an approximation we can use the Hendersen-Hasselbach equation based on the second ionization in Equation 5.

$$pH = pK_a + \log \frac{[base]}{[acid]}$$

This equation can be written as

$$\frac{[base]}{[acid]} = 10^{pH-pK_a}$$

Since our target pH is 8.5 we have $pH - pK_a = -0.5$ and therefore

$$\frac{[base]}{[acid]} = 10^{-0.5}$$

If our target concentration is 0.1 M we can find the needed conjugate [base] and [acid] concentrations

$$\frac{0.1 - x}{x} = 10^{-0.5} = 0.316$$

Therefore, $[base] = 0.024 \text{ M}$ and $[acid] = 0.076 \text{ M}$. Using this calculation we can make the appropriate ratio using three methods. If you have both the acid and conjugate base then method III is the most elegant. Regardless of which method you choose, you will need to check the pH and add a small amount of HCl or NaOH solution to obtain pH 8.5.

The procedures below can be used to make a 0.1 M sodium borate buffer at pH 8.5

Method I

Table 1. Required components to make a 0.1 L of a 100 mM pH 8.5 sodium borate buffer

Component	Mass	Molarity
Boric Acid (H_3BO_3) (MW: 61.83 g/mol)	0.62 g	0.1 M
NaOH (MW: 39.997 g/mol)	0.304 g	0.076 M

1. Prepare 80 mL of dH₂O in a suitable container.
2. Add 0.62 g of Boric Acid (H_3BO_3) to the solution.
3. Add 0.304 g of NaOH to the solution.
4. Add distilled water until volume is 0.1 L.

Method II

Table 2. Required components to make a 0.1 L of a 100 mM pH 8.5 sodium borate buffer

Component	Mass	Molarity
Sodium tetraborate ($Na_2B_4O_7 \cdot 10 H_2O$) (MW: 381.37 g/mol)	3.81 g	0.1 M
HCl (MW: 36.45 g/mol) concentrated acid	0.655 mL	11.6 M

1. Prepare 80 mL of dH₂O in a suitable container.
2. Add 3.81 g of sodium tetraborate decahydrate (Na₂B₄O₇·10 H₂O) to the solution.
3. Add 655 µL of HCl to the solution.
4. Add distilled water until volume is 0.1 L.

Method III

Table 3. Required components to make a 0.1 L of a 100 mM pH 8.5 sodium borate buffer

Component	Mass	Molarity
Sodium tetraborate (Na ₂ B ₄ O ₇ ·10 H ₂ O) (MW: 381.37 g/mol)	0.915 g	0.024 M
Boric Acid (H ₃ BO ₃) (MW: 61.83 g/mol)	0.469 g	0.076 M

1. Prepare 80 mL of dH₂O in a suitable container.
2. Add 0.915 g of sodium tetraborate decahydrate (Na₂B₄O₇·10 H₂O) to the solution.
3. Add 0.469 g of H₃BO₃ to the solution.
4. Add distilled water until volume is 0.1 L.