Buffering Capacity as a Motivating Agent for Buffer Solution Learning

Kevin Husak Carvalho¹, Fabiane Fortes², Tânia Zaleski³, José Roberto Caetano da Rocha⁴

¹(Department of Sciences-Biochemistry/ Federal University of Paraná, Brazil) ^{2, 3, 4}(Collegiate of Biological Sciences / State University of Paraná, Brazil)

Abstract:

Background: In this work, a practical activity was developed, including the construction of graphs, evaluating the power or the buffering capacity of a buffer solution. This concept, although it is presented in several Undergraduate Courses, is considered very abstract and difficult to assimilate by most students. Thus, it is expected that the activity will be a motivating factor in the teaching-learning process of chemical equilibrium, and in turn buffer solution. It was also possible to review the importance of a buffer solution, using blood tissue as the main example. **Materials and Methods**: To visualize the buffering power of the buffer solutions, and after each addition and homogenization, the pH value of the resulting solution obtained with mechanic agitation was determined. The same process was carried out with distilled water, that is, a non-buffer system. **Results**: After all the experiments, graphs of the pH valueswere produced as a function of the volume valuesof the acid solution or the volume of the alkaline solution added to better visualize the result of this process. When evaluating the pH values after adding the volumes of acid or alkaline solutions, the buffering capacity of the buffer solution is clearly perceived, but the results are more visible in the graphs produced. **Key Word**: teaching learning process biochemistry potentiometry.

Key Word: teaching-learning process, biochemistry, potentiometry

Date of Submission: 20-02-2023

Date of Acceptance: 03-03-2023

I. Introduction

Buffer solution is one that can resist significant variations in pH values, when small aliquots of acidic or alkaline solutions are added to the medium [1]. A buffer solution can be obtained by mixing a weak acid and its conjugate base or by mixing a weak base and its conjugate acid, according to the Brönsted-Lowry acid-base theory [2].

The efficiency of a buffer system depends on some factors, but the main one is the concentration of its constituents. The buffering characteristics of a buffer system are maintained as long as the pH value of the original solution does not decrease or increase by one unit, this property being called buffering power or capacity.

The buffer solution plays a decisive role in numerous chemical and biochemical processes [3]. In the human body, blood tissue is an essential buffer system. Numerous biochemical reactions take place in the blood and are responsible for changing pH values, either by adding or subtracting H^+ ions. Evaluating the gaseous exchanges, it is emphasized that the Bohr Effect and the Haldane Effect are two processes that precisely explain the introduction of oxygen gas into the blood tissue and the withdrawal of carbon dioxide produced during the Krebs Cycle [4]. These processes are possible due to the buffer system present in the blood, as they prevent significant changes in pH values. There are four main buffering systems present in blood tissue: the bicarbonate system, the protein system and haemoglobin itself [5].

When evaluating the first system, of the bicarbonate ion, it is noticed that part of the carbon dioxide, introduced due to the metabolic reaction of decomposition of Acetyl-CoA in the Krebs Cycle, is dissolved in the blood tissue according to equation 1 [6].

$$CO_{2(g)} \Leftrightarrow CO_{2(dissolvido)}$$
 Equation 1

When, the reaction of dissolved carbon dioxide with water, present in the blood tissue, occurs, forming the hypothetical carbonic acid, as represented in the chemical equilibrium of Equation 2 [7].

$$CO_{2(dissolvido)} + H_2O_{(\ell)} \Leftrightarrow H_2CO_{3(aq)}$$
 Equation 2

Following this balance, as represented in Equation 3, the hypothetical carbonic acid is ionized with the release of the proton (H^+) and the bicarbonate anion, and this system is an extracellular buffer [8].

$$H_2CO_{3(aq)} \Leftrightarrow H^+_{(aq)} + HCO^-_{3(aq)}K_{a1} = 4,2x10^{-7}$$
 Equation 3

The second blood system is phosphate buffer, considered an intracellular buffer system. In this system there are three ionization processes, but the process that helps in the buffer system of the species is the second (Equation 4), in which the dihydrogenphosphate ion releases an H^+ ion transforming into the hydrogenphosphate ion [9].

$$H_2PO_{4(aq)}^- \Leftrightarrow H_{(aq)}^+ + HPO_{4(aq)}^{2-} K_{a2} = 6.2x10^{-8} \quad Equation \ 4$$

The next chemical species that assists in the blood buffer system is haemoglobin itself, considered an intracellular buffer [10]. Thus, the oxyhaemoglobin present in red blood cells is reduced to haemoglobin, thus releasing oxygen gas molecules in the cells of other tissues of living beings (Equation 5).

$$Hb. O_2 + H^+ \Leftrightarrow Hb. H^+ + O_2$$
 Equation 5

The last blood buffer system is the protein system, in which the amino acids present in protein chains, containing amino or carboxylic functional groups, react by releasing or absorbing protons (H^+) [11]. In these cases, the functional groups act as a weak acid or base, allowing control of the concentration of H^+ introduced into the blood tissue. This system is considered an intracellular buffer [12].

When the protein buffer system has the carboxylic functional group in the side chain, it can release this H^+ ion, thus increasing its concentration, and slightly decreasing the pH values of the blood tissue (Equation 6).

$$Protein - COOH \Leftrightarrow Protein - COO^{-} + H^{+}$$
 Equation 6

When the protein buffer system has an amine functional group in the side chain, it can absorb this H^+ ion, thus decreasing its concentration and slightly raising the pH values of the blood tissue (Equation 7).

$Protein - NH_2 + H^+ \Leftrightarrow Protein - NH_3^+$ Equation 7

The concentration of these chemical species are extremely important to determine the buffering capacity of the blood tissue [13].

This entire metabolic and physiological process makes up the syllabus developed in chemistry and biochemistry classes in undergraduate courses. As it is a topic that involves prior and abstract knowledge, most students consider it difficult to assimilate. In cases like this, there is no formula for the development of an adequate teaching methodology, but it is essential that the teacher review his methods in search of the most suitable means of teaching, reducing the students' difficulty and improving the understanding of that particular topic [14, 15].

Teachers and researchers are unanimous about the importance of carrying out practical activities in the teaching-learning process of natural sciences. This apparent consensus derives from an empirical conception of science and its methods, where the practical character predominates.

However, several authors report a gap between practical activities and their execution [16, 17].

The use of experimentation, especially in the teaching of chemistry and related areas, is an important resource to reduce the difficulties that many students have in understanding the theoretically worked concepts and that teachers face to teach them. Practical or playful activities facilitate the teaching-learning process, enabling the transformation of programmatic content that is difficult to assimilate into something tangible [18]. Thus, these activities become learning motivators [19].

In this context, the present work aimed to develop an activity that facilitated the understanding of the functioning of the buffer system, making the students feel motivated. With practical activity, it is easier to understand that the concepts of capacity or buffering power of a buffer solution are broken when the initial pH value of the buffer solution is increased or decreased by a pH unit [20].

II. Material And Methods

This work was prepared and carried out at the Environmental Impact Assessment Laboratory (Lavima) at the State University of Paraná (Unespar) on the Paranaguá *Campus*.

The equipment used in this experiment was an analytical balance (FA/JA electronic balance operation), magnetic stirrer (78HW-1) and bench-top pH meter model PHS-3E PHTEK.

The activities were divided into two stages, the first was the elaboration and discussion of the methodology and the second was the practical implementation of this method for obtaining and analysing Volume and pH data, consecutively. It was performed on 4 samples in total, one control of distilled water and three of buffer solution, being performed in three replicates to minimize analytical problems. The methodological procedure can be better visualized below (Figure 1).



Figure 1. Schematic sequence of methodological development. Source: Author.

Initially, three volumes of 500mL of phosphate buffer solutions (K_2HPO_4 and KH_2PO_4) were prepared with the following concentrations: 0.01 mol L⁻¹, 0.05 mol L⁻¹ and 0.10 mol L⁻¹, as well as 1L of acidic HCl solution 0.10 mol L⁻¹ and 1L of alkaline NaOH solution 0.10 mol L⁻¹.

Then the analytical tests were performed separately, first with the control sample, with distilled water. Subsequently, all analytical tests were performed with phosphate buffer solution at concentrations of 0.01 mol L^{-1} , 0.05 mol L^{-1} and 0.10 mol L^{-1} . Each of the analytical sequences were performed with three replicates.

For this purpose, 50mL of the sample was transferred separately from each of the buffer solutions and the control sample to a beaker positioned on a magnetic stirrer and successive aliquots of the 0.10 mol L^{-1} HCl solution were added, according to the Table 1. After adding each aliquot of the acidic solution, the pH value of the evaluated control or buffer solution was measured and recorded. This process continued for a different period, until the buffering power of the solution was broken, that is, when the variation in the pH value was 1.0 point in relation to the initial pH value of each of the evaluated solutions. After breaking the buffering power of each of the solution were added to the samples to check the breaking of the buffering.

Evaluated Solution	Volume of acidic or basic solution / μL
Control (distilled water)	50
Phosphate 0,01 mol L ⁻¹	50
Phosphate 0,05 mol L ⁻¹	150
Phosphate 0,10 mol L ⁻¹	300

 Table 1.List of volumes of acid solution or basic solution added to each of the evaluated solutions.

 Source: Author.

For the tests with the addition of aliquots of the alkaline solution, the same procedure was used for the addition of the acid solution, that is, from each of the buffer solutions and the control sample, 50mL of the sample was transferred separately to a beaker positioned in a magnetic stirrer and successive aliquots of the 0.10 mol L^{-1} NaOH solution were added, as shown in Table 1. At the end of the tests, the information obtained experimentally was used to obtain graphs with the ORIGIN software.

III. Result and Discussion

Observing the results of the pH values, obtained during the addition of the acid solution or the alkaline solution, in different volumes of the 0.01 mol L^{-1} buffer solution, presented in Figure 2, it is noticed that the tests carried out in each one of the buffer solutions showed very good repeatability. Similar results were obtained when evaluating the results of the two other buffer solutions.



Figure 2.Graphical representation of the pH values obtained during the addition of the acid solution or alkaline solution, in the 0.01 mol L⁻¹ phosphate buffer solution. Source: Author.

According to the results obtained in the graph (Figure 3) it is noticed that for each sample, the pH values are presented in a standardized and visual way. According to Zabala^[21] and Coll and collaborators^[22], practical activities carried out with a view to the teaching-learning process of students must start from meaningful and functional situations, that is, understand the content in order to internalize it in its reality conveniently, presenting models or activities that help it to look at the content itself.



Figure 3.Graphic representation of the pH values obtained in the control sample and in the phosphate buffer solutions with the addition of aliquots of HCl (0.10 mol L^{-1}) or NaOH (0.10 mol L^{-1}). Source: Author.

In relation to Figure 3, it is also observed that the buffering capacity of the phosphate buffer solution samples is greater in those solutions with higher concentrations. During the process of adding aliquots of the 0.10 mol L^{-1} HCl solution, it was noticed that the pH values were increasingly lower, but very close to the previous values until close to the breakdown of the buffering capacity. During the process of adding aliquots of the 0.10 mol L^{-1} NaOH solution, the results were similar to those observed with HCl, with pH values increasing.

When aliquots of the 0.10 mol L^{-1} HCl solution were added to the control sample, that is, distilled water, the pH values obtained were always lower than the previous one and showed differences much greater than 1.0 point of pH, thus justifying that this solution does not have the power to buffer the medium. The same fact happened when aliquots of the 0.10 mol L^{-1} NaOH solution were added to the control sample, that is, of distilled water, the pH values obtained were always higher than the previous one and presented differences much greater than 1,0 pH point, thus justifying that this solution does not have the power to buffer the medium.

In Table 2 it is possible to observe the volumes of acid solution and alkaline solution necessary to break down the buffering power of each solution. The results are in agreement with Silva and Simoni^[7] who report that the greater the dilution of the buffer solution, the lower its buffering capacity, while the buffer solutions that have a higher concentration demonstrate the opposite. This occurs due to the disturbance of the balance between the chemical species participating in each sample, whether these species are of acidic or alkaline origin [2].

Evaluated Solution	Volume of acidic solution added / µL	Volume of basic solution added / µL
Control (distilled water)	50	50
Phosphate 0,01 mol L ⁻¹	3050	4500
Phosphate 0,05 mol L ⁻¹	16050	17800
Phosphate 0,10 mol L ⁻¹	32600	34700

Table 2. Volumes of acidic or alkaline solution added to break down the buffering power. Source:

 Author.

For applications to be carried out by academics from different undergraduate courses and to facilitate the teaching-learning process, it is suggested that the volumes of acidic or alkaline solution be those shown in Table 3. Thus, less points are obtained to be added in the graphs and in turn applicability in a shorter time. This same table also shows the values of solution that must be added after breaking the buffering capacity.

Evaluated Solution	Volume of acidic or basic solution added / μL		
	Before Breakdown of Buffering Capacity	After Breakdown of Buffering Capacity	
Control (distilled water)	50	100	
Phosphate 0,01 mol L ⁻¹	150	300	
Phosphate 0,05 mol L ⁻¹	500	750	
Phosphate 0,10 mol L ⁻¹	1000	1500	

Table 3. List of volumes of acid solution or basic solution that must be added to each of the evaluated solutions, in future tests. Source: Author.

IV. Conclusion

It is understood that the laboratory practice that discusses the buffering capacity of a buffer solution can help in the assimilation of the syllabus in question, since the visualization during the process facilitates their internalization. Therefore, concepts about concentration, buffering capacity and obtaining graphs can be more easily assimilated by students. With this activity, or with the rereading of it through a conceptual review activity in the classroom, one can clearly see the decrease in the pH values of a solution, when aliquots of acidic solution are added to it, as well as the increase in the pH values of a solution when aliquots of basic solutions are added to it. It is also noticed that the concentration of the buffer solution is directly proportional to the buffering power or capacity.

References

- [1]. Harris, D. C; Lucy, C. A. Quantitative Chemical Analysis. 10th Edition. New York: Macmillan Publishers, 2020, 825p.
- [2]. Marconato, J. C.; Franchetti, S. M. M.; Pedro, R. J., Buffer Solution: An Experimental Proposal Using Low. Química Nova na Escola, v. 20, 2004, p. 59-62.
- [3]. Brown, T. L., Le May JR, H. E., Bursten, B. E., Murphy, C. J.; Woodward, P. M.; Stoltzfus, M. W. Chemistry: The Central Science. 14th Edition.London: Pearson, 2017,1248p.
- [4]. Framer, M. Transição da Respiração Aquática para a Aérea: Efeitos do CO₂ sobre a função da Hemoglobina. Supl. Acta Amazônica, v. 8, n. 4, 1978, p.143-151
- [5]. Fiorucci, A. R.; Hebert, M.; Soares, F. B.; Cavalheiro, E. T. G. The Concept of Buffer Solution. Química Nova na Escola, v.13, 2001, p.18-21.
- [6]. Voet, D; Voet, J. G.; Pratt, C. W.Fundamentals of Biochemistry: Life at the Molecular Level, 5th Edition. New York: Wiley, 2016, 1186p.
- [7]. Silva, C. R.; Simoni, J. A. Buffer Capacity Evaluating A Collaborative Experiment. Química Nova, v. 23, n. 3, 2000, p. 405-409.
- [8]. Brady, J. E.; Senese, F. Chemistry: Matter and its changes. 5th Edition.New York: Wiley, 2007, 1048p.
- [9]. Santos, T. L.; Inaba, J.; Viana, A. G.Avaliação da Capacidade Tamponante da Saliva Uma Aula Prática de Bioquímica. Resumo expandido. 13° CONEX (Conversando sobre Extensão). 2015.
- [10]. Baynes, J. W.; Dominiczak, M. H. Medical Biochemistry. 6th Edition, Netherlands: Elsevier, 2022, 744p.
- [11]. Rodrigues, B. C.; Galembeck, E. Aminoácidos e proteínas: Proposta de atividade prática sob uma abordagem investigativa. Revista Brasileira de Ensino de Bioquímica e Biologia Molecular, v 1, 2011. E1-E10.
- [12]. Nelson, D. L.; Cox, M. M. Lehninger Principles of Biochenistry. 8th Edition. Stuttgart: W. H. Freeman, 2017, 1308p.
- [13]. Marzzoco, A., Torres, B. B. Bioquímica Básica. 3.ed. Rio de Janeiro: Editora Guanabara Koogan S.A., 2007, 386p.
- [14]. Tartif, M. Saberes profissionais dos professores e conhecimentos universitários. Revista Brasileira de Educação, v. 13, 2000. p. 5-24.
 [15]. Rocha, J. R. C. Atividade Lúdica Desenvolvida para Facilitar o Entendimento de como Surgem os Modelos. Holos, v.28, n. 6, p.249-261, 2012.
- [16]. Andrade, M. L. F.; Massabni, V. G. Practical activities development: a challenge to science teachers. Ciência & Educação, v. 17, n. 4,2011, p. 835-854.
- [17]. Coquidé, M. One to look at on the experimentation in the French primary school. Ensaio: pesquisa em educação em ciências, v. 10, n. 1, 2008, p. 118.
- [18]. Moraes, S. R.; Adacheski, D. R.; Cruz, L. H.; Rocha, J. R. C. Investigação da eficácia de aulas teóricas e práticas no processo ensino-aprendizagem de estudantes do 5º ano. Ensino & Pesquisa, v. 14, Suplemento Especial, 2016, p.188-213.
- [19]. Moraes, S. R.; Rocha, J. R. C. Atividade Motivadora para o Aprendizado de Conceitos de Oxidação e Redução. Holos, v.28, n.4, 2012, p.250-258.
- [20]. Skoog, D. A.; West, D. M.; Holler, F. J.; Crouch, S. R. Fundamentals of Analytical Chemistry. 9.ed. United States of America: Saunders College Publishing, 2013, 1072p.
- [21]. Zabala, A. Como trabalhar os conteúdos procedimentais em sala de aula. 2. ed. São Paulo: Artmed Editora, 1999, 194p.
- [22]. Coll, C. Martin, E; Mauri, T; Miras. M; Onrubia, J; Solé, I; Zabala, A. O construtivismo em sala de aula. São Paulo: Editora Ática, 2006, 221p.

Kevin Husak Carvalho, et. al. "Buffering Capacity as a Motivating Agent for Buffer Solution Learning." *IOSR Journal of Biotechnology and Biochemistry (IOSR-JBB)*, 9(1), (2023): pp. 34-39.

DOI: 10.9790/264X-09013439
