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# Preparation of buffer solution lab manual

Standard buffer solutions are solutions with standard pH values used as references for pH measurements and pharmacopoeial tests that require adjustments or maintenance of a specific pH. These solutions can be prepared using methods described below. Special buffer solutions are detailed in sections where their use is specified, such as the microbiological assay of antibiotics or individual monographs indicating the use of such solutions. The reagents needed for preparing standard buffer solutions are listed here. All crystalline reagents except boric acid should be dried at 110° to 120°C for an hour before use. Carbon dioxide-free water should be used when preparing buffer solutions, and this is implied whenever water is mentioned for such preparations. Prepared solutions should be stored in chemically resistant glass-stoppered bottles of alkaline-free glass and used within three months of preparation. Any solution that becomes cloudy or shows signs of deterioration should be discarded. Standard buffer solutions can be prepared over various pH ranges (1.2 to 10.0) by combining hydrochloric acid, sodium hydroxide, and other solutions in specific proportions. The standard pH values listed are considered reproducible within ± 0.02 unit at 25°C. Preparation of Buffer Solutions: 1. Sodium Hydroxide (NaOH) Solution: Mix liquid with carbon dioxide-free water to contain 8.0 g of NaOH in 1000 ml. Note that this solution must be used within one month of preparation. Buffer Solutions: * Hydrochloric Acid Buffer: Combine 50 ml of 0.2M potassium chloride, the specified volume of 0.2M hydrochloric acid, and water to reach a total volume of 200 ml. * Phthalate Buffer: Mix 50.0 ml of 0.2M potassium hydrogen phthalate with the specified volume of 0.2M sodium hydroxide and water to reach a total volume of 200 ml. * Neutralized Phthalate Buffer (Phthalate Buffer): Combine 50.0 ml of 0.2M potassium hydrogen phthalate, the specified volume of 0.2M sodium hydroxide, and water to reach a total volume of 200 ml. * Phosphate Buffer Solution: Mix 50.0 ml of 0.2M potassium dihydrogen phosphate with the specified volume of 0.2M sodium hydroxide and water to reach a total volume of 200 ml. * Alkaline Borate Buffer: Combine 50.0 ml of 0.2M boric acid and potassium chloride, the specified volume of 0.2M sodium hydroxide, and water to reach a total volume of 200 ml. Other Buffer Solutions: * Acetate Buffer pH 2.8: Dissolve 4 g of anhydrous sodium acetate in about 840 ml of water, then add sufficient glacial acetic acid to adjust the pH to 2.8 (about 155 ml) and dilute with water to 1000 ml. * Acetate Buffer pH 3.4: Mix 50 ml of 0.1M sodium acetate with 950 ml of 0.1M acetic acid. * Acetate Buffer pH 3.5: Dissolve 25 g of ammonium acetate in 25 ml of water, then add 38 ml of 7M hydrochloric acid and adjust the pH to 3.5 with either 2M hydrochloric acid or 6M ammonia. Dilute with water to 1000 ml. * Acetate Buffer pH 3.7: Dissolve 10 g of anhydrous sodium acetate in 300 ml of water, then adjust the pH to 3.7 with glacial acetic acid and dilute with water to 1000 ml. Before use, adjust the pH if necessary. * Acetate Buffer pH 4.0: Mix 2.86 ml of glacial acetic acid and 1.0 ml of a 50 percent w/v solution of sodium hydroxide in a 1000 ml volumetric flask, then add water to volume and mix. Adjust the pH if necessary. * Acetate Buffer pH 4.4: Dissolve 136 g of sodium acetate and 77 g of ammonium acetate in water and dilute with water to 1000 ml. Add 250 ml of glacial acetic acid and mix. With water to 100 ml, adjust the pH as needed. Acetate Buffer pH 4.7: Combine 8.4 g sodium acetate and 3.35 ml glacial acetic acid with sufficient water to produce 1000 ml, then adjust the pH if necessary. Acetate Buffer pH 5.0: Mix 13.6 g sodium acetate and 6 ml glacial acetic acid with sufficient water to produce 1000 ml, then adjust the pH as needed. Acetate Buffer pH 5.5: Dissolve 272 g sodium acetate in 500 ml water by heating to 35°C, cool, add 50 ml glacial acetic acid and sufficient water to produce 1000 ml, then adjust the pH if necessary. Acetate Buffer pH 6.0: Combine 100 g ammonium acetate, 4.1 ml glacial acetic acid with 300 ml water, adjust the pH as needed using 10M ammonia or 5 M acetic acid and dilute with water to 500 ml. Acetate Buffer Solution: Mix 14 g potassium acetate and 20.5 ml glacial acetic acid with sufficient water to produce 1000 ml. Barbitone Buffer pH 7.4 Preparation Mix 50 ml of the solution containing 1.944 percent w/v of sodium acetate and 2.946 percent w/v of barbitone sodium with 50.5 ml of 0.1 M hydrochloric acid, add 20 ml of an 8.5 percent w/v solution of sodium chloride and dilute with water to 250 ml. Barbitone Buffer pH 8.6 Preparation Dissolve 1.38 g of barbitone, 8.76 g of barbitone sodium and 0.38 g of calcium lactate in sufficient water to produce 1000 ml. Boric Buffer pH 9.0 Preparation Dissolve 6.20 g of boric acid in 500 ml of water, adjust to pH 9.0 with M sodium hydroxide (about 41.5 ml) and dilute with water to 1000 ml. Buffer Solution pH 2.5 Preparation To 25.0 ml of 0.2 M potassium hydrogen phthalate add 37.0 ml of 0.1 M hydrochloric acid and dilute with sufficient water to produce 100.0 ml. HEPES Buffer Solution pH 7.5 Preparation Dissolve 2.38 g of 2-[4-(hydroxyethyl)piperazin-1-ethanesulphonic acid in about 90 ml of water, adjust the pH to 7.5 with sodium hydroxide solution and dilute to 100 ml with water. Carbonate Buffer pH 9.7 Preparation Dissolve 8.4 g of sodium bicarbonate and 10.6 g of sodium carbonate in sufficient water to produce 500 ml. Chloride Buffer pH 2.0 Preparation Dissolve 6.57 g of potassium chloride in water, add 119.0 ml of 0.1 M hydrochloric acid and dilute with water to 1000 ml. Citrate Buffer Preparation Dissolve 0.5 g of citric acid monohydrate and 0.4 g of dibasic sodium phosphate in sufficient water to produce 1000 ml. Citro-phosphate Buffer pH 5.0 Preparation Mix 48.5 ml of 0.1 M citric acid with sufficient 0.2 M disodium hydrogen phosphate to produce 100 ml. Citro-phosphate Buffer pH 6.0 Preparation Mix 36.8 ml of a 2.1 percent w/v solution of citric acid with 63.2 ml of a 7.15 percent w/v solution of disodium hydrogen phosphate. Citro-phosphate Buffer pH 7.0 Preparation Mix 17.6 ml of a 2.1 percent w/v solution of citric acid with 82.4 ml of a 7.15 percent w/v solution of disodium hydrogen phosphate. Citro-phosphate Buffer pH 7.2 Preparation Mix 13.0 ml of a 2.1 percent w/v solution of citric acid with 87.0 ml of a 7.15 percent w/v solution of disodium hydrogen phosphate. Citro-phosphate Buffer Solution pH 7.6 Preparation Dissolve 1.33 g of citric acid and 67.1 g of disodium hydrogen phosphate in sufficient water to produce 1000 ml. Cupric Sulphate Solution pH 2.0, Buffered Preparation Mix 5.3 ml of 0.2 M hydrochloric acid and 25 ml of 0.2 M potassium chloride, add 4 ml of a 0.393 percent w/v solution of cupric sulfate and dilute to 100ml of water. Cupric Sulphate Solution pH 4.0, Buffered Preparation Dissolve 0.25 g cupric sulfate and 4.5 g of ammonium chloride in sufficient water to produce 250 ml. To prepare Cupric Sulphate Solution: 1. Dissolve 1.522 g of anhydrous disodium hydrogen phosphate in 53.6 ml of water. 2. Add a 2.1% solution of citric acid until the pH is between 5.15 and 5.25. To prepare Diethanolamine Buffer pH 10.0: 1. Dissolve 96.4 g of diethanolamine in 400 ml of water. 2. Add 0.5 ml of an 18.6% w/v solution of magnesium chloride. 3. Adjust the pH to 10.0 with 1 M hydrochloric acid and dilute to 500 ml. To prepare Glycine Buffer pH 11.3: 1. Mix a solution containing 0.75% w/v glycine and 0.58% w/v sodium chloride with an equal volume of 0.1 M sodium hydroxide. 2. Adjust the pH if necessary. To prepare Glycine Buffer Solution: 1. Mix 42 g of sodium bicarbonate and 50 g of potassium bicarbonate with 180 ml of water. 2. Add a solution containing 37.5 g of glycine in 180 ml of water. 3. Dilute with water to 500 ml. To prepare Imidazole Buffer pH 6.5: 1. Dissolve 6.81 g of imidazole and 1.23 g of magnesium sulfate in 752 ml of 0.1 M hydrochloric acid. 2. Adjust the pH if necessary and dilute with water to produce 1000 ml. To prepare Imidazole Buffer pH 7.4: 1. Dissolve 3.40 g of imidazole and 5.84 g of sodium chloride in water. 2. Add 18.6 ml of 1 M hydrochloric acid and dilute with water to produce 1000 ml. To prepare Palladium Chloride Solution, Buffered: 1. Warm 0.5 g of palladium chloride on a water bath with 5 ml of hydrochloric acid. 2. Gradually add hot water in small portions until the solution is complete. 3. Cool and dilute with sufficient water to produce 250.0 ml. To prepare Phosphate-albumin buffered saline pH 7.2: 1. Dissolve 10.75 g of disodium hydrogen phosphate, 7.6 g of sodium chloride, and 10 g of bovine albumin in water. 2. Dilute to 1000.0 ml with the same solvent. To prepare Phosphate Buffer pH 2.0: 1. Dissolve 0.136 g of potassium dihydrogen phosphate in 800 ml of water. 2. Adjust the pH to 2.0 with hydrochloric acid and add sufficient water to produce 1000 ml. To prepare Phosphate Buffer pH 2.5: 1. Dissolve 100 g of potassium dihydrogen phosphate in 800 ml of water. 2. Adjust the pH to 2.5 with hydrochloric acid and add sufficient water Make buffers at specific pH levels by combining and dissolving various chemicals in water to create solutions with desired acidity. For instance: * Create a Phosphate Buffer pH 3.0 by mixing 1.36g of potassium dihydrogen orthophosphate, 2ml of triethylamine, and adjusting the pH with orthophosphoric acid. * Develop a Phosphate Buffer pH 4.9 by dissolving 40g of sodium dihydrogen phosphate and 1.2g of sodium hydroxide in water to achieve a specific pH level. The text outlines various methods for producing buffers at different pH levels, each requiring the combination of distinct chemicals in water to attain the desired acidity. Dissolution Instructions for Various Phosphate Buffers and Saline Solutions #### Phosphate Buffer pH 7.0, 0.1 M Mixed Combine 1.361 g of potassium dihydrogen orthophosphate with water to create 100 ml, then adjust the pH using a 3.5% w/v solution of disodium hydrogen orthophosphate. #### Phosphate Buffer pH 7.5: Dissolution and Adjustment Dissolve 6.8 g of potassium dihydrogen phosphate and 1.56 g of sodium hydroxide in 900 ml of water to adjust the pH to 7.5 with sodium hydroxide solution, then dilute to 1000 ml. #### Phosphate Buffer pH 7.5, 0.2 M Combine 27.2 g of potassium dihydrogen phosphate with 930 ml of water, then adjust the pH to 7.5 using a 0.3% w/v solution of potassium hydroxide. #### Phosphate Buffer pH 7.5, 0.33 M Mixed Dissolve 119.3 g of disodium hydrogen phosphate in sufficient water to create 1000 ml and dissolve 45.36 g of potassium dihydrogen phosphate in sufficient water to produce 1000 ml; mix 85 ml of solution I with 15 ml of solution II and adjust the pH if necessary. #### Phosphate Buffer pH 8.0, 0.02 M Combine 50 ml of 0.2 M potassium dihydrogen phosphate with 46.8 ml of 0.2 M sodium hydroxide and add sufficient water to produce 500 ml. #### Phosphate Buffer, 0.025 M Standard Dissolve 3.40 g of potassium dihydrogen phosphate and 3.55 g of anhydrous disodium hydrogen phosphate in sufficient water to create 1000 ml. #### Phosphate Buffer, 0.05 M Dissolve 6.8 g of potassium dihydrogen orthophosphate in sufficient water to produce 1000 ml. #### Saline, Phosphate-buffered Combine 2.5 g of sodium dihydrogen phosphate, 2.523 g of disodium hydrogen phosphate, and 8.2 g of sodium chloride in sufficient water to create 1000 ml. #### Saline pH 6.4, Phosphate-buffered Dissolve 1.79 g of disodium hydrogen phosphate, 1.36 g of potassium dihydrogen phosphate, and 7.02 g of sodium chloride in sufficient water to produce 1000 ml. #### Saline pH 7.4, Phosphate-buffered Combine 2.38 g of disodium hydrogen phosphate, 0.19 g of potassium dihydrogen phosphate, and 8.0 g of sodium chloride in sufficient water to create 1000 ml; adjust the pH if necessary. #### Tris-Acetate Buffer pH 8.5 Dissolve 0.294 g of calcium chloride and 12.11 g of tris (hydroxymethyl) aminomethane in water, then adjust the pH with 5 M acetic acid and dilute to 1000.0 ml with water. #### Tris-Chloride Buffer pH 7.4 Dissolve 7.27 g of tris (hydroxymethyl) methylamine and 5.27 g of sodium chloride, then adjust the pH if necessary. Tris buffer solution preparation involves dissolving tris (hydroxymethyl) aminomethane in water and adjusting the pH with hydrochloric acid. There are two types of tris buffers, one at pH 7.4 and another at pH 8.1, which require different components and preparation methods. Buffer solutions should be maintained in a controlled environment with optimal moisture levels and protection from contamination, particularly pH-sensitive compounds exposed to light. Storage temperature must also be regulated to ensure stability. Regarding reuse, it's generally advised to utilize freshly prepared buffers for best results. However, if the shelf life of the buffer is known, it can be reused within a specific time frame.

Lab report on preparation of buffer solution.  Preparation buffer solution.  Buffer solution lab.  How are the basic buffer solution prepared.  Buffer solution experiment.