

Jun 19, 2019

Buffer Recipes for Protein Purification

DOI

dx.doi.org/10.17504/protocols.io.4e8gthw

Annie Kwon¹

¹University of California San Francisco



Annie Kwon

University of California San Francisco

Create & collaborate more with a free account

Edit and publish protocols, collaborate in communities, share insights through comments, and track progress with run records.

Create free account

OPEN  ACCESS



DOI: <https://dx.doi.org/10.17504/protocols.io.4e8gthw>

Protocol Citation: Annie Kwon 2019. Buffer Recipes for Protein Purification. **protocols.io**

<https://dx.doi.org/10.17504/protocols.io.4e8gthw>

License: This is an open access protocol distributed under the terms of the **Creative Commons Attribution License**, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

Protocol status: Working

We use this protocol and it's working

Created: June 19, 2019

Last Modified: June 19, 2019

Protocol Integer ID: 24768

Keywords: Buffers, protein purification, buffer recipes for protein purification, buffer recipe

Troubleshooting

Equilibration buffer

1

25 mM HEPES, 100 mM NaCl, pH 7.5

		250 mL	500 mL	1 L	2 L	4 L
	HEPES (MW=238.31)	1.48 94 g	2.97 89 g	5.95 78 g	11.91 55 g	23.8 31 g
	NaCl (MW=58.44)	1.46 1 g	2.92 2 g	5.84 4 g	11.68 8 g	23.3 76 g

Dissolve in water; bring pH up to 7.5; store at 4°C

25 mM HEPES, 500 mM NaCl, pH 7.5

		250 mL	500 mL	1 L	2 L	4 L
	HEPES (MW=238.31)	1.48 94 g	2.97 89 g	5.95 78 g	11.91 55 g	23.8 31 g
	NaCl (MW=58.44)	7.30 5 g	14.61 g	29.2 2 g	58.4 4 g	116.8 8 g

Dissolve in water; bring pH up to 7.5; store at 4°C

Lysis buffer

2 50 mM Na/K phosphate buffer, 500 mM NaCl, 2.5% glycerol, pH 8.0

		250 mL	500 mL	1 L	2 L	4 L
	NaH ₂ PO ₄ • H ₂ O (MW=137.9923)	0.06 90 g	0.13 80 g	0.27 60 g	0.55 20 g	1.103 9 g
	K ₂ HPO ₄ (MW=174.18)	2.09 02 g	4.18 03 g	8.36 07 g	16.7 213 g	33.4 426 g
	NaCl (MW=58.44)	7.30 5 g	14.61 g	29.2 2 g	58.4 4 g	116.8 8 g
	glycerol	6.25 mL	12.5 mL	25 mL	50 mL	100 mL



Dissolve in water; bring pH up to 8.0; store at 4°C

Wash buffer

3 25 mM HEPES, 1M NaCl, 10 mM imidazole, 10% glycerol, pH 7.5

	250 mL	500 mL	1 L	2 L	4 L
HEPES (MW=238.31)	1.4894 g	2.9789 g	5.9578 g	11.9155 g	23.831 g
NaCl (MW=58.44)	14.61 g	29.22 g	58.44 g	116.88 g	233.76 g
imidazole (MW=68.08)	0.1702 g	0.3404 g	0.6808 g	1.3616 g	2.7232 g
glycerol	25 mL	50 mL	100 mL	200 mL	400 mL

Dissolve in water; bring pH up to 7.5; store at 4°C

25 mM HEPES, 1M NaCl, 10 mM imidazole, 10% glycerol, pH 7.5

	250 mL	500 mL	1 L	2 L	4 L
HEPES (MW=238.31)	1.4894 g	2.9789 g	5.9578 g	11.9155 g	23.831 g
NaCl (MW=58.44)	14.61 g	29.22 g	58.44 g	116.88 g	233.76 g
imidazole (MW=68.08)	0.3404 g	0.6808 g	1.3616 g	2.7232 g	5.4464 g
glycerol	25 mL	50 mL	100 mL	200 mL	400 mL

Dissolve in water; bring pH up to 7.5; store at 4°C

Elution buffer

**4** 25 mM HEPES, 100 mM NaCl, 300 mM imidazole, pH 7.5

	250 mL	500 mL	1 L	2 L	4 L
HEPES (MW=238.31)	1.48 94 g	2.97 89 g	5.95 78 g	11.91 55 g	23.8 31 g
NaCl (MW=58.44)	1.46 1 g	2.92 2 g	5.84 4 g	11.68 8 g	23.3 76 g
imidazole (MW=68.08)	5.10 6 g	10.21 2 g	20.4 24 g	40.8 48 g	81.6 96 g

Dissolve in water; bring pH up to 7.5; store at 4°C

25 mM HEPES, 500 mM NaCl, 300 mM imidazole, pH 7.5

	250 mL	500 mL	1 L	2 L	4 L
HEPES (MW=238.31)	1.48 94 g	2.97 89 g	5.95 78 g	11.91 55 g	23.8 31 g
NaCl (MW=58.44)	7.30 5 g	14.61 g	29.2 2 g	58.4 4 g	116.8 8 g
imidazole (MW=68.08)	5.10 6 g	10.21 2 g	20.4 24 g	40.8 48 g	81.6 96 g

Dissolve in water; bring pH up to 7.5; store at 4°C