

Phosphate-Buffered Saline (10× PBS)

Catalog No.
NXB-PBS-10X

Lot: 2025-03-0482

Version: 3.1

Isotonic Cell Biology Buffer — Stock Concentrate

Nexa Biolab Reagent Services • Formulation Date: 14 Mar 2025 • Prepared by: Dr. Clara Vance

Purpose & Scope

This 10× PBS stock concentrate is formulated for use in mammalian cell culture, immunofluorescence, Western blotting, flow cytometry, and general molecular biology applications requiring a precisely buffered isotonic saline solution.

Dilute 1:10 with ultrapure water (Type I, $\geq 18\text{ M}\Omega \cdot \text{cm}$) to obtain standard 1× PBS at physiological ionic strength. The concentrate is stable at room temperature for up to **12 months** and indefinitely at 4 °C when stored in sealed, sterile bottles.

Typical applications:

- Cell washing between staining steps (immunocytochemistry)
- Tissue cryosection rehydration (10 min at RT)
- Antibody and protein dilution buffer
- FACS sheath fluid preparation (filtered, 0.2 μm)
- Equilibration of chromatography columns

Quick Specifications

Property	Value
Form	Liquid
Appearance	Clear, colorless
pH (10×)	6.8–7.0
pH (1×)	7.2–7.4
Osmolality	$\approx 2800\text{ mOsm/kg}$
Sterility	Non-sterile
Endotoxin	$< 0.1\text{ EU/mL}^\dagger$
Grade	Molecular Biology

[†] LAL assay; sterile-filtered lots only.

Validated Reagent

Nexa Internal Standard QC-44B

Passed: 14 Mar 2025

Component Table — Final Concentrations in 10× PBS

Component	MW (g/mol)	Conc. (10×)	Amount / L	Supplier Cat. #	Role
Sodium chloride (NaCl)	58.44	1.37 M	80.1 g	NXR-NaCl-500	Ionic strength
Potassium chloride (KCl)	74.55	27 mM	2.0 g	NXR-KCl-100	Ionic balance
Disodium hydrogen phosphate (Na ₂ HPO ₄)	141.96	100 mM	14.4 g	NXR-DSP-250	Buffer base
Potassium dihydrogen phosphate (KH ₂ PO ₄)	136.09	18 mM	2.4 g	NXR-KDP-250	Buffer acid
Ultrapure water (Type I)	—	—	q.s. to 1 L	NXR-H2O-MQ	Solvent
After 1:10 dilution (1× PBS):		NaCl 137 mM • KCl 2.7 mM • Na ₂ HPO ₄ 10 mM • KH ₂ PO ₄ 1.8 mM			

Preparation Protocol

1. Weigh all solid components to ± 0.01 g accuracy using an analytical balance (Nexa PrecisioScale A200).
2. Dissolve salts in ~ 800 mL ultrapure water in a 2 L borosilicate glass beaker with magnetic stirring.
3. Stir at room temperature ($20-25^{\circ}\text{C}$) for ≥ 15 min until fully dissolved.
4. Adjust pH to 6.8–7.0 (see pH Adjustment note).
5. Bring total volume to exactly 1.0 L with ultrapure water.
6. Transfer to clean 1 L glass bottles; cap tightly.
7. For sterile lots: filter through $0.22\ \mu\text{m}$ PES membrane under laminar flow (Nexa FlowGuard II BSC).
8. Label with lot number, date, preparer, and expiry.
9. Log in LIMS under protocol PBS-10X-SOP-v3.

pH Adjustment Note

The 10 $\times$ concentrate is titrated to pH 6.8–7.0. Upon dilution to 1 $\times$, the pH rises slightly to the target physiological range of 7.2–7.4.

Adjustment agents:

- Raise pH: 1 M NaOH (dropwise, with stirring)
- Lower pH: 1 M HCl (dropwise, with stirring)

Measurement: Calibrate pH electrode immediately before use with NIST-traceable buffers at pH 4.0, 7.0, and 10.0. Allow reading to stabilise (> 30 s) before recording. Temperature-compensate to 25°C . Cross-check with indicator strips (range 6–8) as a secondary method.

! Do not use autoclave-sterilised lots for pH-sensitive applications. Autoclaving may shift pH by ± 0.3 units.

Storage Conditions

	4°C (primary)
	$\text{RT} \leq 25^{\circ}\text{C}$
	12 months RT
	36 months 4°C
	Sealed container
	Keep from light

 Cold chain not required for short-term shipping (< 7 days)

Discard if precipitate forms and does not redissolve upon warming to 37°C for 10 min.

Safety & Handling

-  Not classified as hazardous (GHS).
-  Eye contact: flush with water ≥ 15 min.
-  Skin: wash thoroughly with soap and water.
-  Inhalation: move to fresh air.
-  Disposal: drain to sewer (check local regulations).

PPE required:

- ★ Nitrile gloves (standard lab)
- ★ Safety goggles / splash shield
- ★ Lab coat

SDS available on Nexa internal portal:
safety.nexabiolab.int/sds/NXB-PBS-10X

QC Release Criteria

Test	Spec.
pH (10 $\times$)	6.8–7.0
pH (1 $\times$)	7.2–7.4
Osmolality	2750–2850
Appearance	Clear
Sterility	Pass (sterile)
Endotoxin	< 0.1 EU/mL
Conductivity	≤ 33 mS/cm

All tests performed per Nexa SOP-QC-44B. Certificate of Analysis available on request. Lot retention sample stored at -20°C .

Troubleshooting Guide

Observation	Probable Cause	Corrective Action	Prevention
White precipitate at 4 °C	Phosphate salt crystallisation due to over-concentration	Warm to 37 °C and mix gently; if unresolved, discard lot	Verify final volume during prep
pH out of range after dilution	Incorrect concentration of stock components	Recheck component weights; re-prepare stock	Calibrate balance quarterly
Microbial growth (turbidity)	Contaminated source water or container	Discard; re-filter or autoclave container	Use sterile vessels; work in BSC
Unexpected cell toxicity	Endotoxin contamination or wrong ionic strength	Submit for LAL testing; verify osmolality	Use depyrogenated glassware

Common Variants & Modifications

Variant	Modification	Application	Notes
PBS-T (PBST)	+ 0.05 % Tween-20	Western blot wash buffer; ELISA block & wash	Add Tween-20 after pH adj.
Ca ²⁺ /Mg ²⁺ PBS	+ 0.1 mM CaCl ₂ , 1 mM MgCl ₂	Cell adhesion assays; organ culture systems	Prepare fresh; Ca/Mg precipitate
DPBS (Dulbecco)	No KCl, KH ₂ PO ₄	Routine cell washing (ATCC formulation)	See Nexa cat. NXB-DPBS
Immunostaining PBS	+ 0.1 % BSA, 0.02 % NaN ₃	Antibody dilution for IHC/IF	NaN ₃ : toxic; handle in fume hood
PBS-EDTA	+ 2 mM EDTA (pH adjusted)	Cell detachment; flow cytometry prep	EDTA chelates Ca ²⁺ , Mg ²⁺

References & Related Documents

- Nexa Biolab SOP PBS-10X-SOP-v3 (internal)
- Nexa QC Standard: SOP-QC-44B
- Formulation basis: Green & Sambrook (2012), *Molecular Cloning*, 4th ed., CSHL Press (adapted, no KCl variant: Appendix 1, A1.14)
- ASTM D1193 Standard Specification for Reagent Water (Type I)
- ISO 11135:2014 Sterilisation of health-care products
- Nexa LIMS protocol ref: LIMS-2025-PBS-0482

Revision History

Ver.	Date	Change Summary
1.0	03 Jun 2021	Initial release
2.0	11 Jan 2023	Added Ca ²⁺ /Mg ²⁺ variant; revised QC table
3.0	02 Sep 2024	Endotoxin limit revised; LIMS integration added
3.1	14 Mar 2025	Troubleshooting section added; minor typographic corrections