

# EzPBS(-) Instruction Manual

August 15th, 2016 Ver. 2

## 1. Safety precaution

Before using the product, read this instruction manual thoroughly at first. Do not start the operation until you understand the contents of manual. Also, this document describes only method used for specified purpose with this product. Do not use this product for any purpose or by any method other than described here. If you use this product for any purpose or by any method other than described in this manual, you will be held responsible for any necessary safety measures as operator.

## 2. Application

This product is phosphate buffered saline (PBS(-)) not including calcium and magnesium. PBS(-) has equal osmotic pressure to in vivo and consists of the same ion composition. It is the most generally used for washing and suspending cells because it is nontoxic to cells. On the other hand, it is also useful buffer for protein preparation and biochemical analysis. Especially it is used as blocking reagent of western blotting and immunostaining of cell/tissue, and wash buffer as well as used for preparation of antibody diluent.

## 3. Package

Product name	Volume	Package
<b>EzPBS(-)</b>	1000 mL	1 bottle

## 4. Components

Product name	Main components
<b>EzPBS(-)</b>	1.37 M NaCl, 27 mM KCl, 100 mM Na <sub>2</sub> HPO <sub>4</sub> , 17.6 mM KH <sub>2</sub> PO <sub>4</sub> /pH7.4 (This is 10x concentrated stock solution.)

This product doesn't include notifiable materials exceeding to regulated amount for exclusion decided by PRTR Law, Poisonous and Deleterious Substances Control Act and Industrial Safety and Health Law.

## 5. Storage

- Keep **EzPBS(-)** at room temperature, away from direct sun light. If the package is unopened, it is stable until mentioned expiration date (1 year from production date).
- **EzPBS(-)** is sterilized, but if various germs mix after opening the package, the sterilized condition may not be maintained.

- **EzPBS(-)** doesn't include a preservative. Do not add a blocking reagent to the stock solution.

## 6. Disposal method

- Follow the disposal method decided by the organization you belong to.
- Material of bottle (Main body, Lid): Polypropylene

## 7. Necessary things other than this product

- Distilled water
- **EzTween**, **EzBlock series** etc.

## 8. Precautions for use

- This is 10x concentrated stock solution. Dilute it for use in accordance with the usage.
- **EzPBS(-)** is sterilized, but if various germs mix after opening the package, the sterilized condition may not be maintained.
- **EzPBS(-)** doesn't include a preservative. Be careful of contamination at the time of opening.
- This product doesn't include sodium azide inhibiting the activity of HRP.

## 9. Usage

### A. In the case of use for suspension of cell culture and washing

1. Open **EzPBS(-)** in a clean bench under the germfree condition.
2. Dilute it 10 times with sterilized distilled water under the germfree condition. For example, add 90 mL distilled water to 10 mL **EzPBS(-)** and mix them.

※If germfree technique is hard to perform, sterilize **EzPBS(-)** with autoclave (121°C, 20 min) after diluting it 10 times with distilled water, and use it for cell culture etc.

※Perform same preparation even in the case of use without DNase. (This product is not free of RNase)

### B. In the case of use for washing of western blotting and immunostaining of cell/tissue.

1. Dilute **EzPBS(-)** 10 times with distilled water. For example, add 90 mL distilled water to 10 mL **EzPBS(-)** and mix them.
- Add a surfactant such as **EzTween**, TritonX-100

and so on as needed.

※Add a 0.02-0.5% surfactant at final concentration. According to ATTO standard protocol, it is 0.1% at final concentration.

※It is possible to add a surfactant to stock solution in advance and preserve it (0.2-5% at final concentration). Add a surfactant to stock solution so as to be 10 times more concentrated than at using for working solution.

※In the case of using at high salt concentration, add salt such as NaCl as needed.

C. In the case of use for antibody wash

1. Dilute **EzPBS(-)** 10 times with distilled water. For example, add 90 mL distilled water to 10 mL **EzPBS(-)** and mix them.

2. Add a surfactant such as Tween 20, TritonX-100 and so on as needed.

※Add a 0.02-0.5% surfactant at final concentration.

※It is possible to add a surfactant to stock solution in advance and preserve it (0.2-5% at final solution). Add a surfactant to stock solution so as to be 10 times more concentrated than at using for working solution.

3. Add a blocking solution (**AE-1475,1476,1477 EzBlock series**) as needed.

※When you add **EzBlock series**, mix working solution of both **EzBlock series** and **EzPBS(-)**. In the case of use for a antibody wash in general, it is used at 1/5-1/10 concentration of blocking solution. Mix **EzBlock series** and **EzPBS(-)** in the ratio of 1:5-10.

※Gelatin, skim milk and so can be used as blocking solution instead of **EzBlock series**.



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