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# **5% Blocking Buffer**

Cat. No: E-IR-R107

Size: 5 mL / 10 mL / 50 mL

Cat	Products	5 mL	10 mL	50 mL	Storage
E-IR-R107	5% Blocking Buffer	5 mL	10 mL	50 mL	2~8 ℃

#### Introduction

5% Blocking Buffer is a common western blotting and immuno-histochemical Blocking Buffer, which is suitable for horseradish peroxidase (HRP), alkaline phosphatase (AP) and biotin labeled second antibody. The preservatives in this product has not affect the activity of HRP and AP, and it will not interfere with the detection of HRP or AP labeled secondary antibody. At the same time, the product does not contain biotin and will not interfere with biotin based detection.

This product is a Ready-to-Use product. It can be directly used to block the membrane and immunohistochemistry without any additional reagent.

### **Experimental Procedure**

- 1. Western Blotting Experiment Blocking
  - 1) After membrane transfer, wash the imprinted membrane with Western washing solution for 1~2 min.
  - 2) Pour 5% Blocking Buffer into a plate or other appropriate containers to ensure that the Blocking Buffer can fully cover the membrane.

Tip: Conventional western method, a  $6.6~\text{cm} \times 8.5~\text{cm}$  membrane is recommended to use about 10 ml of Blocking Buffer.

- 3) Clamp one corner of the membrane with flat tweezers, place the membrane in the Blocking Buffer, immerse the membrane in the Blocking Buffer completely, put it on the horizontal shaking table for about 1 h.
- 4) The blocked membrane can be used in the follow-up experiments such as primary antibody incubation according to the steps of western blotting.
- 2. Immunohistochemistry Experiment Blocking
  - 1) Dewaxe and hydrate the tissue slices.
  - 2) If necessary, heat repair the antigens or digestion the slices according to the situation of antigen and antibody.
  - 3) Add 3% H2O2 (It is recommended to use Elabscience® E-IR-R115) to the slice and incubate for 10 min at room temperature to inactivate the endogenous enzyme.
  - 4) Wash the slice with PBS for 3 min/time, 3 times.
  - 5) Add 5% Blocking Buffer to the slice and block at room temperature for 30 min. Then discard the Blocking Buffer.
  - 6) The blocked slice can be used in the follow-up experiments such as primary antibody incubation according to the steps of immunohistochemistry.

## **Storage**



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Store at 2~8 ℃ for 12 months. Avoid of freezing.

#### **Cautions**

- 1. Usually we incubate with the Blocking Buffer at RT for 1 h. It can also be incubated at  $4\,\mathrm{C}$  overnight.
- 2. There is no Blocking Buffer suitable for all experimental systems, for some special experiments, it may be necessary to consider using other more suitable Blocking Buffer according to the specific situation.
- 3. Please keep the Western blotting membrane or immunohistochemical slice moist, otherwise it may produce abnormal background.
- 4. This product couldn't be used for clinical diagnosis or treatment, food or medicine, and can't be stored in residence.
- 5. For your safety and health, please wear the lab coat and disposable gloves before the experiments.