

Product Datasheet

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BCIP/NBT 1 Component Membrane Alkaline Phosphatase Substrate (Purple Reaction Product)

Prod. No.: B390

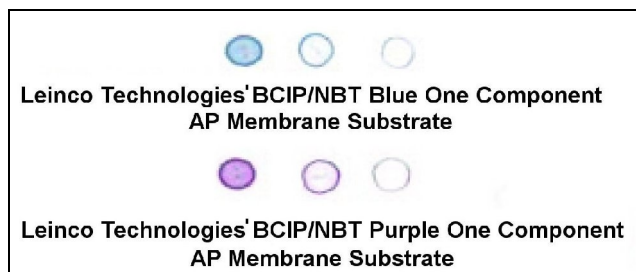
Pkg. Size: 100 ml, 500 ml, 1 L

Storage: 2 – 25 °C *Detailed storage instructions below.*

Description

This BCIP/NBT 1 component buffered alkaline phosphatase substrate (5-bromo-4-chloro-3-indolyl-phosphate and nitroblue tetrazolium) is a precipitating substrate used with the enzyme alkaline phosphatase designed for various membrane immunoassays but not recommended for microwell or immunohistochemical applications. Initially, the unreacted substrate should be colorless to light yellow in appearance. When this substrate system is reacted with alkaline phosphatase, an insoluble, permanent dark purple reaction product is obtained.

Leinco Technologies' BCIP/NBT 1 Component Membrane Substrate is a highly sensitive, high performance formulation that produces a high signal to noise ratio with low membrane background. The outstanding shelf life of at least thirty six months for the BCIP/NBT 1 Component Membrane Substrate makes this reagent ideal for long term use of the same manufacturing lot.



Directions for Product Use

BCIP/NBT 1 Component Membrane Substrate is a ready to use solution that needs no preparation or dilution. This product should be at room temperature (25°C) before use. Pour the necessary amount of substrate onto the blocked alkaline phosphatase conjugate labeled membrane to completely cover the membrane. The substrate will react with sites on the membrane containing phosphatase, producing an insoluble purple reaction product. The reaction should be monitored and read before the background color becomes too intense which may result in diminished contrast between the positive and background staining. To stop the reaction, rinse membrane with reagent quality water. Some wash solutions containing detergents may reduce color intensities. Dilution of the substrate is not recommended. To reduce the intensity of a reaction, it is recommended that the antibodies or conjugates be diluted.

Storage and Stability

The high quality of the substrate can be preserved by storing at temperatures from 2 – 25°C. When properly stored, BCIP/NBT 1 Component Membrane Substrate is stable for a minimum of 3 years from the manufactured date. The substrate should not be frozen and should be protected from direct light by storing in amber bottles. Only high quality amber glass and plastic products should be used for storing aliquots.

Reported Applications

Western, Northern and Southern blot procedures



Products are for research use only. Not for use in diagnostic or therapeutic procedures.

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Country of Origin

USA

Related Products

UltraAvidin™-Alk. Phos. ([Leinco Prod. No.: A108](#))

Streptavidin- Alk. Phos. ([Leinco Prod. No.: S208](#))

Goat Anti-Mouse IgG (H&L)- Alk. Phos. ([Leinco Prod. No.: M290](#))

Goat Anti-Human IgG (H&L)-Alk. Phos. ([Leinco Prod. No.: H627](#))

Goat Anti-Rat IgG (H&L)-Alk. Phos. ([Leinco Prod. No.: R1216](#))

Goat Anti-Rabbit IgG (H&L)-Alk. Phos. ([Leinco Prod. No.: R1191](#))

Goat Anti-Armenian Hamster IgG (H&L)-Alk. Phos. ([Leinco Prod. No.: A130](#))

Troubleshooting Guide

Problem	Cause	Solution
Too much background signal observed	BCIP/NBT substrate was left on the membrane too long	Decrease the amount of time the BCIP/NBT substrate is on the membrane
	Too much primary antibody used	Decrease the amount of primary antibody used and wash the membrane sufficiently after the primary antibody incubation
	Too much secondary antibody used	Decrease the amount of secondary antibody used
Nonspecific bands show up on the membrane	Too much primary antibody used	Decrease the amount of primary antibody used and wash the membrane sufficiently after the primary antibody incubation
	Too much secondary antibody used	Decrease the amount of secondary antibody used
Signal disappears from membrane	Membrane not stored correctly	Store the membrane dry in plastic bag in the dark
No signal is observed on the membrane	Low amounts of specific signal present	Expose the membrane to BCIP/NBT substrate for a longer period of time. Include positive control(s) during analysis
	Insufficient primary antibody used	Use more primary antibody
	Insufficient secondary antibody used	Use more secondary antibody
	Sample degraded	Add protease inhibitors to original sample before running a gel

References

1. C. Waltenbaugh., *et al.* (2002). *Int. Immunol.* **14**:1239.
2. C. Waltenbaugh., *et al.* (2001). *Alcohol Clin. Exp. Res.* **25**:1221.



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