Beacle, Inc.

Signal Booster Neo

Protein-free immuno-reaction enhancing solution

Instruction

Beacle, Inc.
KYOTO JAPAN

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Cautions

1. Research use only. Do not use for medical purpose.
2. Do not dilute or add other agents in Signal Booster Neo solution to get the best result.

(1) Introduction

Signal Booster Neo is, like Signal Booster, an enhancer of antigen-antibody reaction. In Western blotting and ELISA, researchers often experiences weak signal or high background. Signal Booster Neo improves these problems by just using it as antibody diluents. Due to the principle of working mechanism, Signal Booster Neo can be used in many assay systems that use antigen-antibody reaction.

Signal Booster Neo is, unlike Signal Booster, consisted of all chemical components and contains no proteins. So this product is useful for assay systems that have to be avoided protein contamination.

How Signal Booster Neo Works

Signal Booster Neo contains polymers. One polymer, by changing the physicochemical properties of antigen and antibody, enhances the mutual accessibility, and facilitate the specific reaction. The other polymer reduces non-specific binding of antibody. Thus, Signal Booster Neo enhances the antigen-antibody reaction while reducing background.

● Features of Signal Booster Neo●

1. High signal with low background

Signal Booster Neo enhances the antigen-antibody reaction. Comparing with the method using detergent-containing buffer, several to over 10-fold stronger signal can be obtained while the background level is low. Thus, you can get much higher S/N ratio than usual method.

2. Effective for saving antibody usage and time of reaction time

Because higher signal can be obtained using Signal Booster Neo, you can reduce the amount of antibody used and the time required for reactions.

3. Can be used for many reactions

Signal Booster Neo can be used not only for Western blotting and ELISA, but also for other assay systems using antigen-antibody reactions. In addition, Signal Booster Neo does not affect activities of HRP or ALP, and can be used for assay systems using these enzymes.

4. Easy to use

Signal Booster Neo is one solution format and formulated as to Ready to Use. Just exchange your dilution buffer of antibodies to Signal Booster Neo solution.

(2) Products

Signal Booster family has following products. This manual applies all these products.

Product #	Product name	Content
BCL-SBN-01	Signal Booster Neo 250 mL	250 mL
BCL-SBN-02	Signal Booster Neo 500 mL	500 mL

(3) Related products

Some of related products are listed below. To make 100% protein-free assay system we recommend c-Block.

Categories	Product #	Product name	Content
enhancer	BCL-125	Signal Booster 250mL set	250 mL X 2
Blocking solutions	BCL-BKCW-01	c-Block-w (western)	500 mL
	BCL-BKEE-01	c-Block-w (ELISA)	500 mL
Western	BCL-EZQ23	Easy-WESTERN-II Quick, full set	50 tests

Blocking solutions for ELISA are designed for long term storage of precoated plate.

(4) Principle of usage of Signal Booster Neo

- Signal Booster Neo is one solution format. Just use the same solution for the dilution of the 1st and 2nd antibody. No change is required for the other assay protocol. For details see the later instructions.
- Do not dilute the Signal Booster Neo solution. The solution is optimized. Dilution causes loss of Signal Booster Neo's ability.
- Signal Booster Neo can be used in Western blotting, antibody sandwich ELISA with either the 1st or 2nd antibody-labeled type, antigen sandwich ELISA.

(5) Western Blotting (WB)

WB is a method to detect proteins by specific antibodies. Usually, proteins are separated by SDS-PAGE and transferred to membrane made by nitrocellulose or PVDF. The way to use Signal Booster Neo in WB is described below.

1) SDS-PAGE and transfer of protein to PVDF membrane should be done by usual method.
2) Blocking and the washing should be done by usual method.
3) Dilute the 1st antibody with Signal Booster Neo. The dilution factor is influenced by many factors, such as antibody species, amount of antigen, etc. Though you can reduce antibody concentration by using Signal Booster Neo, we recommend performing a pre-test to determine the best antibody concentration.
4) Dilute the 2st antibody with Signal Booster Neo. The best dilution factor is influenced by many factors, such as antibody species, amount of antigen, etc. Refer to the supplier's instruction to determine the best antibody concentration.
5) For visualization, many users use HRP-, or AP-labeled antibody. In both cases, please watch the strength of staining or luminescence and stop the reaction. Longer reaction gives you high background or appearance of extra band.

(6) ELISA

ELISA is a method to determine the amount of antigen or antibody in samples by using labeled antigen or antibody. The sandwich ELISA is most widely used, where antigen sample is applied on solid phase antibody and bound antigen is reacted by HRP-labeled detection antibody. The way to use Signal Booster Neo in the sandwich ELISAs are described below.

1) Antibody attachment (solid phase), blocking and the washing procedure should be done by usual method.
2) Dilute the antigen with PBS-T. Pour appropriate amount of these diluents into each well, mix and incubate for appropriate time. Signal Booster Neo may be used for antigen dilution in order to facilitate reaction of antigen and solid phase antibody.
3) Dilute the HRP-labeled detection antibody with Signal Booster Neo. The best dilution factor is influenced by many factors, such as antibody species, amount of antigen, etc. Refer to the supplier's instruction to determine the best antibody concentration.
4) For substrate reaction, follow the ordinary method.
5) In some case 1st antibody and HRP-labeled 2nd antibody system is used. In such a case use Signal Booster Neo for dilution of both antibodies.

・In other ELISA system, use Signal Booster Neo for reaction buffer for antigen-antibody.

(7) Trouble shooting

Trouble	Cause and resolutions
Western blotting	
Weak signal	1. Low antigen conc.: Use higher antigen conc.
	2. Low antibody conc.: Survey best antibody conc.
	3. Not enough transfer: Use higher current or longer transfer time.
	4. Blocking too strong: Do not use long time blocking.
	5. Too much transfer: When using nitrocellose, proteins pass though the membrane by strong transfer manipulation. Check the procedure or exchange membrane to PVDF.
Partial whitening (lumines.)	6. Too much antigen or antibody: Over signaling often suppress the luminescence and cause partial whitening of a band. Control the amount of antigen or antibody concentration.
Too many extra-bands	7. Too much higher antibody conc.: Higher antibody conc. often causes non-specific signaling. Control the antibody conc.
	8. Too much antigen: Higher antigen often causes non-specific signaling. Control the amount of antigen.
	9. Not enough blocking: Some antigen and antibody have preference of blocking agents, change the blocking agents or check the blocking conditions.
	10. Not enough washing: Increase the number and time of washing.
High background	11. High antibody conc. or too long incubation: Reduce the antibody conc. or shorten the incubation time.
ELISA	
Weak signal	1. Too low antigen or antibody conc.: Increase the concentrations.
Too strong signal	2. Too high antigen or antibody conc.: Check the antigen and antibody concentration by performing the titration.
	3. Too long incubation: shorten the incubation time.
High background	4. Too high antigen or antibody conc.: check antigen and antibody cocn.
	5. Not enough blocking: Some antigen and antibody preference of blocking agents, change the blocking agents or check the blocking conditions.
	6. Not enough washing or too much washing: check the number and time of washing.

(8) Contact Information

E-mail: technical-support@beacle.com

http://www.beacle.com

[manufactured and distributed by]

Beacle, Inc.

10-25 Kamikazan-Sakajiri, Yamashina-Ku,
Kyoto, 607-8465 Japan