



DATA SHEET

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> PDS Standard

PDS (Protein Detection System) is a highly sensitive enhanced substrate for detecting horseradish peroxidase (HRP) on immunoblots. The extremely intense output signal enables detection of picogram amounts of antigen.

The sensitivity, intensity and duration of the signal allow for detection of HRP using photographic or other imaging methods. Blots can also be repeatedly exposed to film to obtain optimal results or stripped of the immunodetection reagents and re probed.

cat. no.	amount	note
STS-E500	250ml + 250ml	peroxide solution and enhancer solution

FOR RESEARCH USE ONLY

SHIPPING

Shipped at room temperature.

STORAGE

Store at 4°C

SHELF LIFE

12 months

FORM

liquid

RECOMMENDED PROCEDURE

peroxide solution and enhancer solution at a 1:1 ratio.

Optimization and notes:

- For best results, it is essential to OPTIMIZE all components of the system including sample amount, primary and secondary antibody concentration, and the choice of membrane and blocking reagents.
- Because no blocking reagent is optimal for all systems, empirical testing is essential to determine the appropriate blocking buffer for each Western blot system. Determining the proper blocking buffer can help increase sensitivity and prevent nonspecific signal caused by cross-reactivity between the antibody and the blocking reagent. Furthermore, when switching from one substrate to another, a diminished signal or increased background sometimes results because the blocking buffer was not optimal for the new system.
- Use sufficient volumes of wash buffer, blocking buffer, antibody solution and substrate working solution to cover blot and ensure that it never becomes dry. Using large blocking and wash buffer volumes minimizes nonspecific signal.
- For optimal results, use a shaking platform during incubation steps
- Add Tween®-20 (final concentration of 0.1-0.2%) to the blocking buffer and all diluted antibody solutions to minimize nonspecific signal.
- Do not use sodium azide as a preservative for buffers. Sodium azide is an inhibitor of HRP and could interfere with this system.
- Do not handle membrane with bare hands. Always wear gloves or use clean forceps
- Exposure to the sun or any other intense light can harm the substrate. For best results avoid prolonged exposure to any intense light. Short-term exposure to typical laboratory lighting will not harm the working solution.



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PROCEDURE SUMMARY

Primary Antibody Dilution Range: 1:1000 - 1:5000

Secondary Antibody Dilution Range: 1:10000 - 1:100000

- Prepare the working solution by mixing the two substrate components (peroxide solution and enhancer solution) at a 1:1 ratio.

Note: Exposure to the sun or any other intense light can harm the substrate. For best results avoid prolonged exposure to any intense light. Short-term exposure to typical laboratory lighting will not harm the working solution.

- Incubate blot 1 min. in the working solution.
- Drain excess reagent. Cover blot with clear plastic wrap.
- Expose blot to X-ray film.

Detailed Western Blotting Procedure:

- Remove blot from the apparatus and block nonspecific sites with blocking buffer* for 60 minutes at room temperature (RT) with shaking.
- Rinse with wash buffer* and add the appropriate primary antibody diluted in blocking buffer. Incubate for 1-2 hours with shaking. If desired, blots may be incubated with primary antibody overnight at 2-8°C.
- Rinse the membrane in wash buffer with shaking for ≥ 10 minutes. Replace wash buffer at least 3 times. Increasing the wash buffer volume and/or the number of washes may help reduce background.
- Incubate blot with the appropriate HRP-conjugate secondary antibody diluted in blocking buffer for 1-2 hour at RT with shaking.
- Repeat washes to remove non-bound HRP-conjugate.
- Prepare working solution by mixing equal parts of peroxide and enhancer solution. Use 0.1 ml working solution per cm² of membrane.
- Incubate blot in the working solution.
- Drain excess reagent. Cover blot with clear plastic wrap. Use an absorbent tissue to remove excess liquid and to carefully press out any bubbles from between the blot and surface of the membrane protector.
- Place the protected membrane in a film cassette with the protein side facing up. Turn off all lights except those appropriate for film exposure (e.g., a red safelight).
- Carefully place a piece of film on top of the membrane. A recommended first exposure time is 30 seconds. Exposure time may be varied to achieve optimal results. Enhanced or pre-flashed film is not necessary.

Note: The exposure time may be varied to achieve optimal results. If the signal is too intense, reduce exposure time or optimize the system by decreasing the antigen and/or antibody concentrations.

- Light emission is most intense during the first 5-30 minutes after substrate incubation. Light emission will decrease with time.
- Blots may be stripped and reprobed if necessary.

*Supported buffers:

blocking buffer A : 1xTBS, 0,1% Tween®-20, 3% low fat dry milk

blocking buffer B : 1xPBS, 0,2% Tween®-20, 5% low fat dry milk

washing buffer A : 1xTBS, 0,1% Tween®-20

washing buffer B : 1xPBS, 0,2% Tween®-20

*For research only, not for resale