

UltraScience Femto Western Substrate

15 MAR 2023

Catalog Number	Size	Reaction(s)
CCH365-B100ML	50 ml x 2	Sufficient for over 25 mini-gel size membranes.
CCH365-004ML	2 ml x 2	Sufficient for over one mini-gel size membrane.

Storage Conditions

Stable for up to 24 months at 4°C, do not freeze it.

Shipping Condition

Ship at 4°C, beware of shipping in any condition beneath 0°C.

Description

The **UltraScience Femto Western Substrate**, as a luminol-based enhanced chemiluminescent substrate, is sensitive and compatible with conducting immunoblots with horseradish peroxidase (HRP) – conjugated secondary antibodies. The mid femtogram to high femtogram detection of antigen is enabled by UltraScience Femto Western Substrate's excellent sensitivity and long signal duration. Further, its long chemiluminescent signal duration makes both digital and film-based imaging possible without any loss of the signal. Appropriate primary and secondary antibody dilutions are suggested for attaining optimal signal intensity and duration.

- **No optimization required.** Switching to the UltraScience Femto Western Substrate from other brands, such as Pierce and GE Healthcare, does not require optimization or protocol changes.
- **High degree of sensitivity and enhanced chemiluminescence duration.** UltraScience Femto Western Substrate enables an accurate mid femtogram to high femtogram detection of protein on the same immunoblot after a single exposure.
- **Optimized for use with PVDF and nitrocellulose membranes.**
- **Compatible with Western Blotting Markers.**
- **Optimized for film- and CCD-based imaging.**

Kit Content(s)

Catalog Number	Size
CCH365-B050MLA	50 ml x 1
CCH365-B050MLB	50 ml x 1

Required materials but not provided

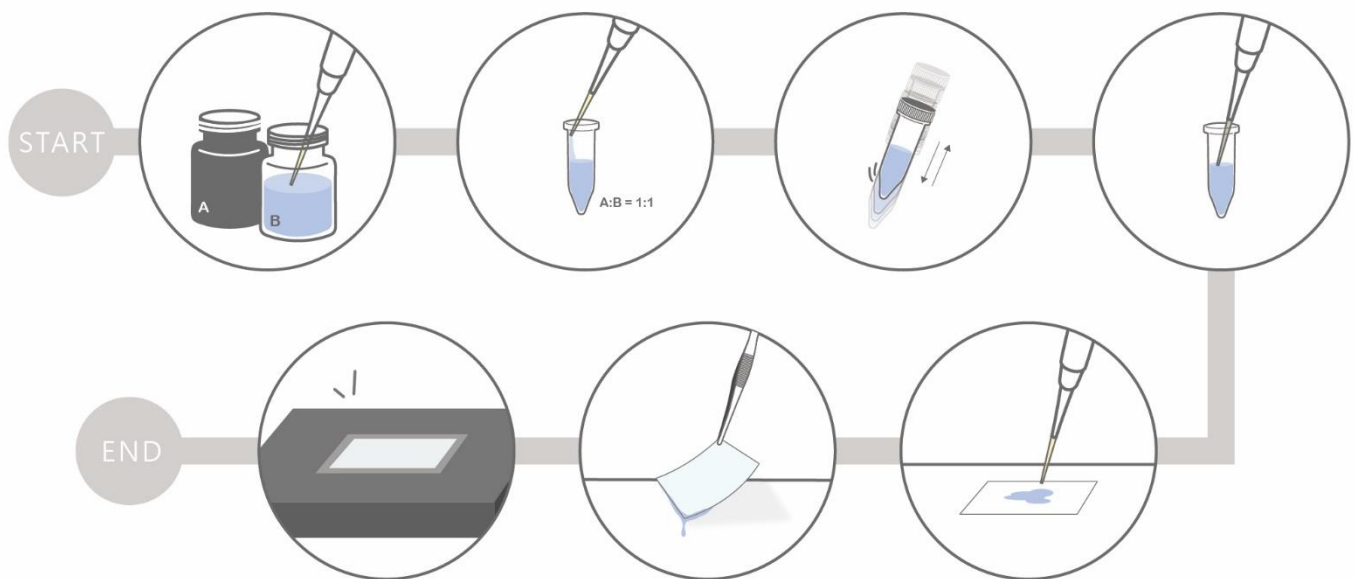
- A compatible Chemiluminescence or X-ray Imaging Systems
- A plastic sheet protector or plastic wrap to prevent the membrane from drying

Instrument Compatibility

This western substrate is compatible with the majority of commercially available Chemiluminescence and X-ray Imaging Systems.

Reaction Setup

1. Keep the membrane moist in the wash buffer while preparing the substrate mixture. Please ensure the membrane does not dry out during the subsequent steps.
2. Mix Luminol solution and Peroxide Solution in a 1:1 ratio, and thoroughly agitate the chemiluminescent substrate solution well for preparing the 0.1 ml of solution / cm² of membrane.
 - For a mini-sized membrane (7 x 8.5 cm), 4 ml of solution is sufficient.
 - For a midi-sized membrane (8.5 x 13.5 cm), 10 ml of solution is sufficient.
3. Place the membrane with the protein side up on a clear and level surface or in a clean container.
4. Remove the membrane from the chemiluminescent substrate solution and drain off excessive solution.
5. Place the membrane in a plastic sheet protector or in plastic wrap to prevent the membrane from drying.
6. Image the membrane with a digital imager or by exposing to the X-ray film.





Important notes

UltraScience ECL substrates series is compatible with the use from low picogram to low-femtogram level detections. Please kindly refer to the ECL selection guide of UltraScience Western substrate as the below table.

Bio-Helix Western Substrates	Advantages for you	Sensitivity	Compare Performance to
UltraScience Pico Plus Western Substrate CCH321-B100ML	Best value of abundant protein detection and best sensitivity among entry-level western substrate	low picogram or high femtogram	-Thermo Scientific™ Pierce ECL Substrate -Thermo Scientific™ SuperSignal™ West Pico PLUS -CYANAGEN™ WESTAR™ ETA C 2.0 -Advansta™ WesternBright™ ECL -Expenden™ LumiBlue ECL Extra
UltraScience Pico Ultra Western Substrate CCH345-B100ML	Better choice when seeking low abundance proteins, over 30 times sensitivity than UltraScience Pico Plus western substrate.	low picogram to mid femtogram	-Millipore™ Immobilon™ Western Substrate -FUJIFILM Wako™ ImmunoStar™ Zeta -GE Healthcare Amersham™ ECL™ Prime -Advansta™ WesternBright™ Quantum™ -Expenden™ LumiBlue™ ECL Extended -CYANAGEN™ WESTAR™ ETA C ULTRA 2.0 -Thermo Scientific™ SuperSignal™ West DURA
UltraScience Femto Western Substrate CCH365-B100ML UltraScience Femto Plus Western Substrate CCH375-B100ML	Born to seek , seeking most less abundance proteins in your Western Blot, even low femtograms.	mid femtogram to low femtogram	-FUJIFILM™ Wako™ ImmunoStar™ LD -GeneTex™ Trident™ femto -Thermo Scientific™ SuperSignal™ West Femto -Advansta™ WesternBright™ Sirius™ -Expenden™ LumiBlue™ ECL Extreme -CYANAGEN™ WESTAR™ SUPERNOVA™ -GE Healthcare™ SELECT

Troubleshooting

Problem	Cause	Solution
High Background	Overconcentrated primary or secondary antibody	*Decrease the antibody concentration.
		*Perform a dot blot to optimize the concentration.
	Insufficient wash	*Increase the frequency or duration.
	Incomplete blocking	*Decrease the antibody concentration.
		*Perform a dot blot to optimize the concentration.
No Reaction or Weak Signal	Insufficient antigen binding	*Decrease antibody concentration. *Optimize blocking reagents for achieving a balance between sensitivity and specificity.
	Poor antibody binding to the antigen	*Optimize detergent used for antibodies. *Increase the antibody incubation time.
No Reaction or Weak Signal	Proteins washed from the membrane during assay	*Reduce the number or intensity of wash
	Insufficient reagent volume	*Apply additional volumes of antibody blocking reagent, or wash solution.