

Store at 4°C

#12757

SignalFire™ Elite ECL Reagent



Support: +1-978-867-2388 (U.S.)
cellsignal.com/support

Orders: 877-616-2355 (U.S.)
orders@cellsignal.com

For Research Use Only. Not for Use in Diagnostic Procedures.

| Product Includes | Product # | Quantity |
|---------------------------------|-----------|----------|
| 12757P | | |
| SignalFire™ Elite ECL Reagent A | 94855P | 10 mL |
| SignalFire™ Elite ECL Reagent B | 17816P | 10 mL |
| 12757S | | |
| SignalFire™ Elite ECL Reagent A | 94855S | 50 mL |
| SignalFire™ Elite ECL Reagent B | 17816S | 50 mL |

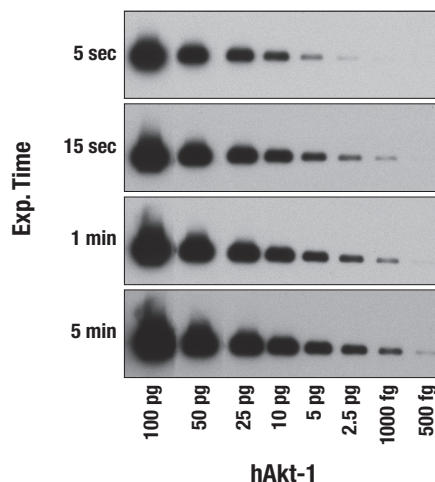
Description: SignalFire™ Elite ECL Reagent from Cell Signaling Technology (CST) is an ultra sensitive chemiluminescent substrate capable of detecting femtogram amounts of protein by western blot analysis. SignalFire™ Elite ECL Reagent is compatible with both film and digital imaging systems. The extremely intense signal output allows detection of very low abundance proteins, conservation of reagents, and short exposure times.

SignalFire™ Elite ECL Reagent requires approximately ten-fold less Anti-rabbit IgG, HRP-linked Antibody #7074 or Anti-mouse IgG, HRP-linked Antibody #7076 than traditional ECL reagents. Limiting the amount of HRP exposed to the membrane prevents high background, oversaturation of the target protein signal, or false negative results. Other HRP-conjugated antibodies, including HRP-conjugated primary and anti-biotin-HRP antibodies, should be diluted similarly. Dilution of secondary antibody from alternative vendors may need to be optimized. Titration of lysate and primary antibody concentration is recommended to achieve optimal signal-to-noise ratio.

Light emission is maximal immediately after exposure of the substrate to HRP and continues for up to 24 hours.

Background: Chemiluminescence systems have emerged as the best all-around method for western blot detection. They eliminate the hazards associated with radioactive materials and toxic chromogenic substrates. The speed and sensitivity of these methods are unequalled by traditional alternatives, and because results are generated on film, it is possible to record and store data permanently. Blots detected with chemiluminescent methods are easily stripped for subsequent reprobing with additional antibodies. HRP-conjugated secondary antibodies are utilized in conjunction with specific chemiluminescent substrates to generate the light signal. HRP conjugates have a very high turnover rate, yielding good sensitivity with short reaction times.

Solutions and Reagents: Each SignalFire™ Elite ECL Reagent A (#94855) and Reagent B (#17816) is a 2X concentrate; there is no need to further dilute in water when the two reagents are combined.



SignalFire™ Plus Elite Reagent is sensitive enough to detect femtogram levels of protein. Western blot analysis of diluted recombinant human Akt-1 protein using Akt (pan) (C67E7) Rabbit mAb #4691.

Storage: Store at 4°C. This product is stable for 12 months. DO NOT store Reagent A (#94855) and Reagent B (#17816) pre-mixed. Reagents A and B should be combined just prior to exposing membranes.

Directions for Use:

- Wash membrane-bound HRP (antibody conjugate) at least three times for 5 minutes in TBS/T. It is very important to thoroughly wash the membrane prior to substrate incubation.
- Prepare 1X SignalFire™ Elite ECL Reagent by diluting one part 2X Reagent A (#94855) and one part 2X Reagent B (#17816) (e.g. for 10 mL, add 5 mL Reagent A and 5 mL Reagent B). Mix well.
- Incubate substrate with membrane for 1 minute, remove excess solution (membrane remains wet), wrap in plastic and expose to X-ray film.

*Avoid repeated exposure to skin (see enclosed Material Safety Data Sheet or refer to our website for further information).

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