Cat # SL100309 Store at 4 °C

LumiGOLD™ ECL Western Blotting Detection Kit

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☐ **Standard Pack** **250 ml**

☐ **Large Pack** **1000 ml**

This product is for laboratory research ONLY and not for diagnostic use

Description:

LumiGOLD™ ECL Kit is a complete kit with ready-to-use reagents for chemiluminescent detection of immobilized proteins (Western blotting) or immobilized nucleic acids (Southern or Northern), conjugated with HRP directly or indirectly.

The use of enhanced chemiluminescence was introduced by Thorpe and Kricka (1,2). In the presence of hydrogen peroxide (H_2O_2), Horseradish peroxidase (HRP) catalyzes the oxidation of cyclic diacylhydrazides, such as luminol. Immediately following the oxidation, the luminol is in an excited state (intermediate reaction product), which decays to the ground state by emitting light. Strong enhancement of the light emission is produced by enhancers, such as phenolic compounds. Using this method, it is possible to detect membrane immobilized specific antigens, or sequences of nucleic acids, labeled directly with HRP or indirectly with HRP-labeled antibodies/streptavidin.

Quick Protocol:

1. Mix an equal volume of LumiGOLD™ ECL Kit Reagent A and Reagent B to give sufficient solution to cover the membrane (0.1ml/cm2). Let the detection mix equilibrate for 2 minutes at room temperature.
2. Drain the excess buffer from the washed blots. Do not let the membrane dry out. Add the detection mix directly to the blot (protein side up). Incubate for 2 minutes at room temperature.
3. Drain off excess detection mix and wrap the membrane in saran wrap. Gently remove air pockets.
4. Place the blots, protein side up, in the film cassette. Switch off the lights and use red safety light. Place a sheet of film on the blot, close the cassette and expose for 30-90 seconds.
5. Replace the exposed film with a new one, close the cassette and develop the first exposed film.
6. Expose the second film for a suitable time according to the signal intensity on the first film.
7. If signal intensity was too high, wait up to 30 minutes before re-exposing.

For detailed protocol, please Email us at info@signagen.com to request the full detailed protocol.

References:

(1) Thorpe, G.H.G. and Kricka, L.J., *Methods in Enzymolo*gy, **1**33:331-353 (1986)
(2) Thorpe, G.H.G., Kricka, L.J., Moseley, S.B. and Whitehead, T.P., *Clin. Che*m., **31(8):**1335-1341 (1985)
(3) Riko, I., et al, *Analytical Biochemist*ry, **231:**170-174 (1995)

Storage: Upon arrival store this product at 4 ° C. Product shipped at cold-pack filled foam box.