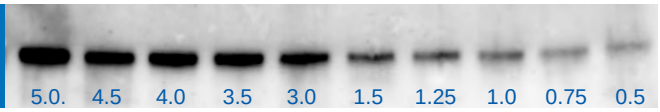
SERVA*Light* HRP Chemiluminescence Kits

A New Family of Substrates

SERVA*Light* HRP chemiluminescence kits

SERVA offers four different types of highly sensitive ready-to-use kits for chemiluminescence detection of membrane bound antigens (Western Blot) or nucleic acid sequences (Southern and Northern Blot), labeled directly with Horseradish Peroxidase (HRP) or indirectly with HRP-conjugated antibodies/streptavidin.

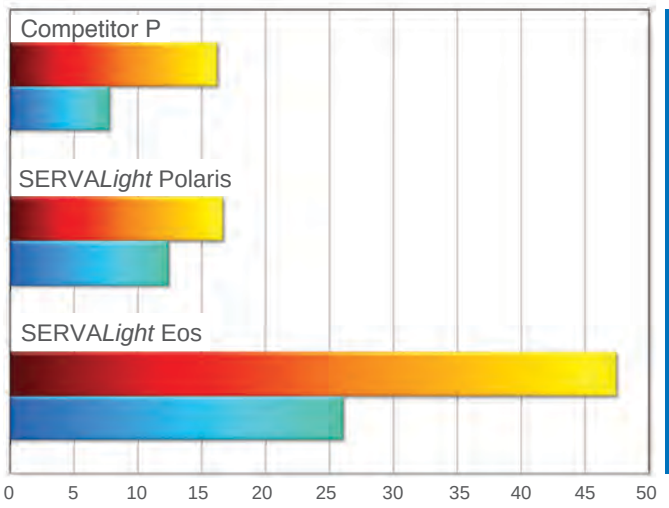
- **Easy to use** – simply mix the two components, a luminol/enhancer solution and a stabilized peroxide solution in a one-to-one ratio.
- **Excellent stability** – at least one year stable when stored at room temperature
- **Safe to use** – no hazardous components
- **Extended signal duration** – all substrates show long light emission, but signal duration is optimized for SERVA*Light* EosUltra with an outstanding light emission for 18 – 20 hours at a very high signal
- **Versatile signal detection** – detection can be done by film or CCD cameras
- **Economical** - save money and precious antibodies due to high dilution of antibodies

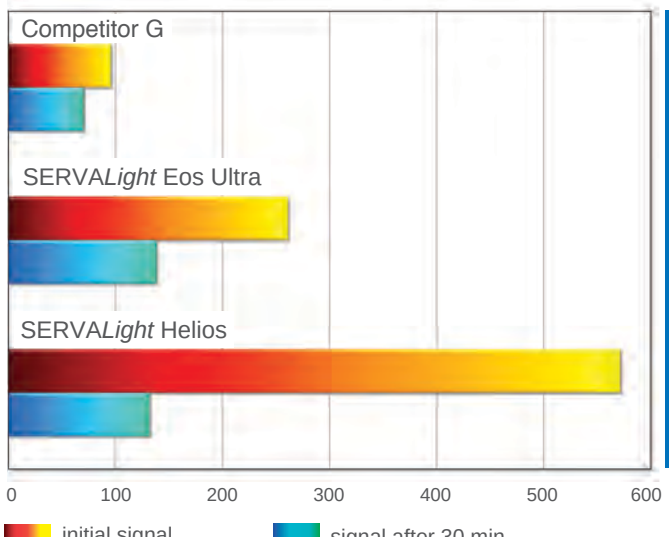


Human Transferrin was diluted (5 to 0.5 ng) and electrophoresis was performed. The gel was transferred to PVDF membranes, blocked and incubated with 1:20,000 rabbit anti-transferrin. After washing, the membranes were incubated with 1:100,000 of HRP-conjugated goat anti-rabbit antibody. The membrane was washed again and then incubated with SERVA*Light* EOSUltra. Exposure time was 300 sec.

Signal intensity over time of SERVA*Light* HRP chemiluminescence kits

The following diagrams show a comparison of the relative light output (chemiluminescence signal intensity) produced by various HRP substrates upon addition of a given amount of HRP. The yellow-red bar corresponds to the intial signal, while the bluish bar represents the remaining signal after 30 min.





Data obtained by Cyanagen Srl.

	SERVA*Light* Polaris	SERVA*Light* Eos	SERVA*Light* EosUltra	SERVA*Light* Helios
Lower detection limit	Low picogram	High femtogram	Mid femtogram	Low femtogram
Signal duration	5 – 6 h	11 – 12 h	18 – 20 h	7 – 8 h
Primary detection method	Autoradiography film	Imaging equipment or autoradiography film	Imaging equipment or autoradiography film	Imaging equipment or autoradiography film
Suggested antibody dilution (1 mg/ml)	Primary: 1:500 – 1:5000 Secondary: 1:20.000 – 1:100.000	Primary: 1:1000 – 1:15.000 Secondary: 1:25.000 – 1:150.000	Primary: 1:5000 – 1:50.000 Secondary: 1:50.000 – 1:250.000	Primary: 1:5000 – 1:100.000 Secondary: 1:100.000 – 1:500.000
Working solution stability	24 h at RT	24 h at RT	8 h at RT	8 h at RT
Stock solution stability	>1 year at RT	>1 year at RT	>1 year at RT	>1 year at RT
Recommended membrane	Nitrocellulose or PVDF	Nitrocellulose or PVDF	Nitrocellulose or PVDF	Nitrocellulose or PVDF

The development of the new HRP chemiluminescence kits*

The advantages over other detection methods have allowed chemiluminescence to become the method of choice for Western Blots in most protein laboratories. The long signal duration allows multiple exposures to obtain the optimum image. Reprobing of the blot for visualization of another protein or for optimizing the detection of the first protein, a large linear response range allowing detection and quantitation of a broad range of protein concentrations and, most important, the greatest sensitivity are further advantages of chemiluminescence detection.

Luminol is one of the most widely used chemiluminescence reagents. Peroxidase-catalyzed oxidation of luminol by peroxide creates an excited state product called 3-aminophthalate. By transit to a lower energy state the product emits a weak flash of light at 425 nm. By adding an enhancer (electron transfer mediator) the flash signal is converted into a glow and signal intensity and duration are greatly improved [1, 2]. The light output can be further largely increased by addition of a suitable acylation catalyst as a secondary enhancer [3, 4, 5, 6]. In addition the background light emission is greatly reduced.

* Produced by Cyanagen Srl. Cyanagen Srl is subject of US and EU patent application number US7803573; EP1962095; US7855287; EP1950207, together with other equivalent granted patents and patent applications in other countries.

References

1. Kricka, L. J. (2000) Methods Enzymol. 305, 370–390
2. Heindl, D. and Josel, H. P. (1997) Non-radioactive Analysis of Biomolecules, 258–261, Springer, Berlin
3. Marzocchi, E., et al. (2008), Anal. Biochem., 377, 189–194
4. Vdonvenko, M. M., et al. (2009) Anal. Biochem., 392, 54–58
5. Vdonvenko, M. M., et al. (2010) Biotechnology Journal, 5(8), 886–890
6. Vdonvenko, M. M., et al. (2010) Journal of Immunological Methods, 362 (1–2), 127–130

Ordering information

Product	Size	Cat. No.
SERVA*Light* POLARIS CL HRP WB Substrate Kit	100 ml	42584.01
	250 ml	42584.02
	500 ml	42584.03
SERVA*Light* EOS CL HRP WB Substrate Kit	50 ml	42585.01
	250 ml	42585.02
	500 ml	42585.03
SERVA*Light* EOSUltra CL HRP WB Substrate Kit	20 ml	42586.01
	100 ml	42586.02
	200 ml	42586.03
SERVA*Light* HELIOS CL HRP WB Substrate Kit	20 ml	42587.01
	100 ml	42587.02
	200 ml	42587.03

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