CYANAGEN

Reagents for Molecular Biology

NEW

THE RIGHT LIGHT

ELISTAR

ECL substrates for CLIA

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www.cyanagen.com

About us

Cyanagen is a biotech company located in Bologna, dedicated to research, development and production of reagents for molecular diagnostic since 2003 and one of the leading companies in the field of reagents for Western blotting and Elisa.
The main product lines are focused on chemiluminescence and fluorescent dyes for biological analysis, genomics, proteomics and chemical sensors.
They are based on Cyanagen internationally patented technologies and achieve outstanding performance in terms of sensitivity and stability.
The products are extremely versatile and perfectly suited to the latest analytical instrumentation. These products are also available as OEM.

Cyanagen s.r.l. has a certified Quality System

ISO 9001-2008 QUALITY CERTIFIED

All ELISTAR substrates are protected by **US7803573, EP1962095, US7855287, EP1950207, US2012009603 (A1), CA2742025, EP2405016**, foreign equivalents and pending patents.

Product manual

ELISTAR

ECL substrates for CLIA

ELISTAR IS INTENDED FOR RESEARCH USE ONLY AND SHALL NOT BE USED IN ANY CLINICAL PROCEDURES OR FOR DIAGNOSTIC PURPOSES.

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1. Introduction

ELISTAR is Cyanagen's product line of HRP ECL substrates developed for chemiluminescent ELISA applications. These luminol-based\luminol derivative substrates offer a broad linear dynamic range and a superior signal-to-noise ratio.
Our proprietary technology enables fine-tuning of signal intensity in order to obtain a customized assay sensitivity best suited to your particular application.
ELISTAR product line constitutes a perfect component of every chemiluminescent ELISA kit.

Storage/expiry

One year at room temperature (max. 25°C).

ELISTAR product line

ELISTAR	NOVA 2.0	ANTARES	ETA C ULTRA 2.0	SUPERNOVA	HYPERNOVA
Assay sensitivity	Sensitive	Very Sensitive	Super Sensitive	Ultra Sensitive	Extremely Sensitive
Signal duration	Medium	Extended	Extended	Short	Short
Analyte abundance	High	Medium	Low	Ultra Low	Extremely Low

2. Components and other materials required

Kit components

- **Solution A:** Luminol-based\luminol-derivative\enhancer solution (amber bottle)
- **Solution B:** Peroxide solution (white bottle)

Other materials required

- 96-well opaque microplate
  Capture antibody diluted to 1-10 µg/mL in 50 mM carbonate/bicarbonate pH 9,6
- Blocking buffer (Tris buffered saline (TBS) or Phosphate buffered saline (PBS) with 0.05-0.1% Tween-20 and 1-5% of a blocking reagent, such as bovine serum albumin (BSA), gelatin, casein, non-fat dry milk).
- Washing buffer (TBS or PBS with 0.05-0.1%).
- Primary antibody at 0.05-0.5 µg/mL
- HRP conjugate at 10-20 ng/mL

3. Optimization

- Optimization of the antigen, antibody and HRP-conjugate concentrations may be required. It is important to test a range of parameters, usually by completing a checkerboard dilution series to test various conditions in systematic manner.
- Optimization of coating buffer is suggested to find the optimal condition of binding reactivity in order to enhance the specific signal.
- Optimization of blocking and washing buffers are recommended to reduce background noise and enhance specific signal.
- Buffers, temperature, and humidity must be kept constant between and within experiments in order to produce standardized results.
- Do not use azide as a preservative because azide is a known inhibitor of HRP.

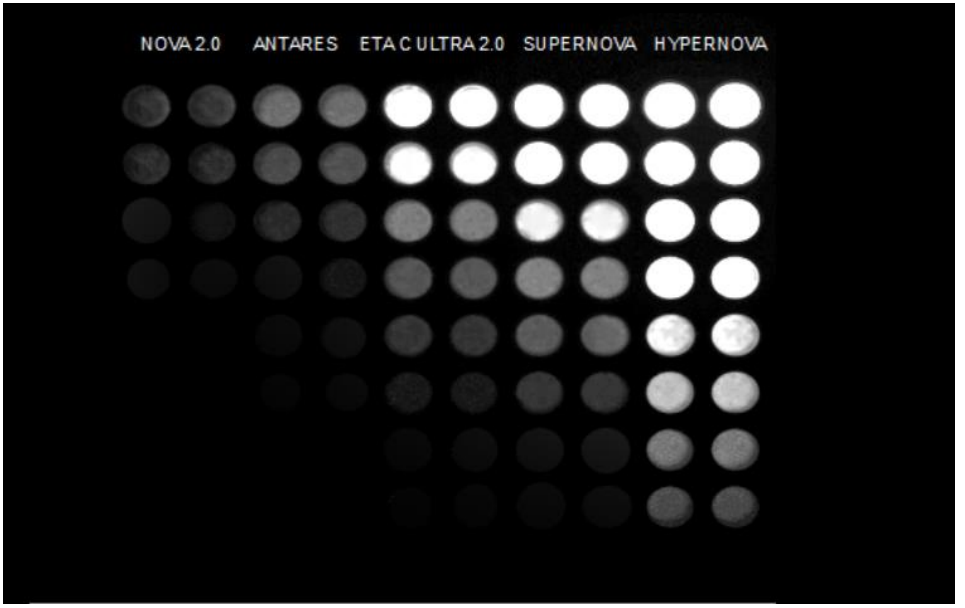
4. Procedure

- Coat, block and wash microplate wells according to standard ELISA procedures.
- Prepare ELISTAR Working Solution by mixing equal parts of Reagent A and B.
- Note: DO NOT expose Working Solution to intense light. Store in an amber bottle.
- Add 100-150 µl of ELISTAR Working Solution to each microplate well.
- Mix liquid in wells for 1 minute using a microplate stirrer.
- Use a microplate luminometer to measure relative light units (RLU) at 425 nm, between 1-5 minutes after adding the Working Solution. Longer exposure may result in decreased signal intensity.

5. Troubleshooting

Symptom	***Possible Cause***	***Solution***
High background	High HRP-conjugate concentration	Decrease HRP-conjugate concentration.
	Incomplete washing	Wash plate wells thoroughly and blot 96-well plate on paper towels to empty wells, prior to the incubation with the chemiluminescent working solution
		Increase the number of washing cycles
	Protein in blocking solution may react nonspecifically with HRP-conjugate	Change the blocking solution.
Signal disappears quickly	High HRP-conjugate concentration has exhausted the substrate prematurely	Decrease HRP-conjugate concentration.
		Decrease antigen concentration.
Weak or no signal	Antibody concentration is too low	Use a higher sensitivity HRP detection substrate.
		Increase Ab concentration.
		Increase antigen concentration.

6. Selection Guide



ELISTAR substrates (Cyanagen) were used for detection of 2-fold dilutions of HRP in a 96-well plate.
Imager: ImageQuant™ LAS 4000 (GE Healthcare).

product	cat#	Assay sensitivity	volume
ELISTAR NOVA 2.0	XLSE077,0100	Sensitive	2X50 mL
ELISTAR ANTARES	XLSE0144,0100	Very Sensitive	2X50 mL
ELISTAR ETA C ULTRA 2.0	XLSE079,0100	Super Sensitive	2X50 mL
ELISTAR SUPERNOVA	XLSE2,0100	Ultra Sensitive	2X50 mL
ELISTAR HYPERNOVA	XLSE150,0020	Extremely Sensitive	2X10 mL

For further information, visit **www.cyanagen.com**
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