

Chromascan

For reliable epi-fluorescence
only available with our new imager IRIS

- New imaging system,
- New technologies,
- New possibilities



UVITEC Cambridge is proud to introduce its new Chromascan, an **unprecedented technology** integrated into our latest imaging system, the Alliance IRIS. This innovative imager is dedicated to all your **chemiluminescence and epi-fluorescent Western blots**, making it your ideal companion for a wide range of scientific applications. IRIS not only presents the **highest optic sensitivity** with an aperture of f/0.75 but also unprecedented features for **unmatched precision** and detection.

■ Why is Chromascan revolutionary?

Our unique Chromascan concept combines the power of the **Laser Emitting Diode (LED) illumination** with the reliability of a **scan-like** system. Our new innovative technology offers both **homogeneous illumination and unmatched sensitivity** for your epifluorescent Western blots applications, representing a significant leap forward in molecular imaging capabilities.



■ The concept of epi-fluorescence and the importance of multiplexing

Epi-fluorescence has been widely used as a technique for recognition of specific proteins. It involves proteins being prepared with a secondary antibody conjugated with a specific fluorophore (**Fig. 1a.**), that will emit a signal only when excited by a certain light source corresponding to its wavelength (**Fig. 1b.**).

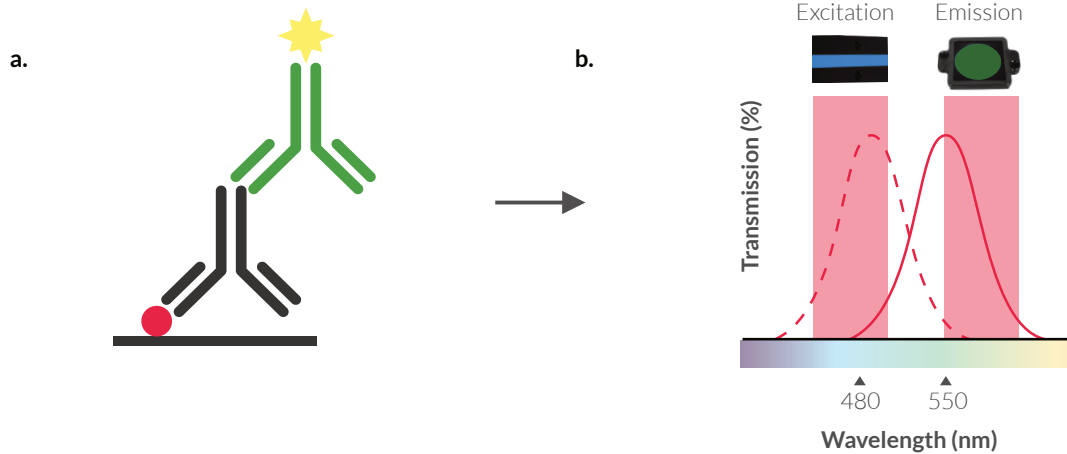


Figure 1. The concept of epi-fluorescence

1a. Visual representation of antibody (red) being detected by primary (black) and secondary antibody (green) which emits light (yellow) at a specific wavelength. **1b.** Diagram of excitation and emission source for corresponding fluorophore wavelength

Multiplexing in epi-fluorescence is a technique that allows detection and analysis of multiple distinct targets using different fluorophores (**Fig. 2.**).

This technique significantly enhances experimental efficiency by reducing the time and resources needed compared to sequential analysis. There is no need to re-probe and re-strip a membrane like those in a chemiluminescent Western blot.

Multiplexing enables comprehensive data collection from complex biological systems, providing a more detailed understanding of interactions and processes. Additionally, it improves the accuracy of results by allowing direct comparison within the same experimental context. This makes it invaluable in fields like molecular biology, clinical diagnostics, and drug discovery. Its power : image multiple proteins on the same Western blot.

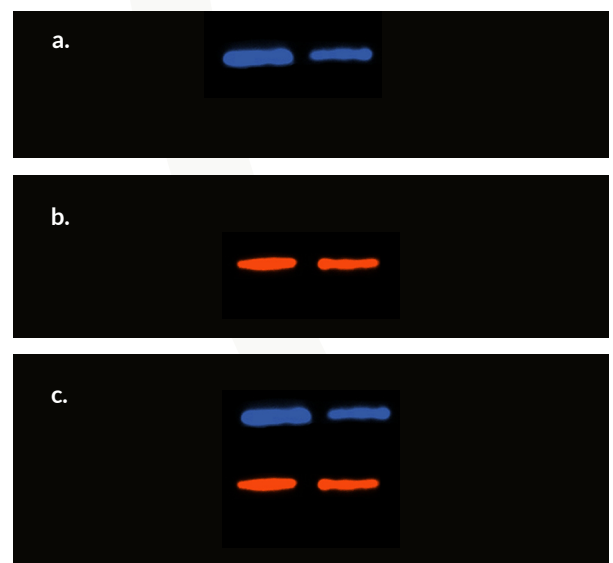


Figure 2. Multiplexed Cy2 and Cy5

2a. Protein X conjugated with Cy2 with blue excitation module (480 nm) and blue emission filter (550nm) **2b.** Protein Y conjugated with Cy5 with red excitation module (650nm) and red emission filter (700nm) **2c.** Multiplexed image of protein X (Cy2) and protein Y (Cy5)

■ Chromascan : consistent illumination increasing your data accuracy

Traditional illumination methods such as **spot LED** is interesting in epifluorescence for its stability overtime and its energy efficiency (Fig. 3.), but **its limitation lies in delivering uniform light over your Western blot**, making your quantification unprecise (1).

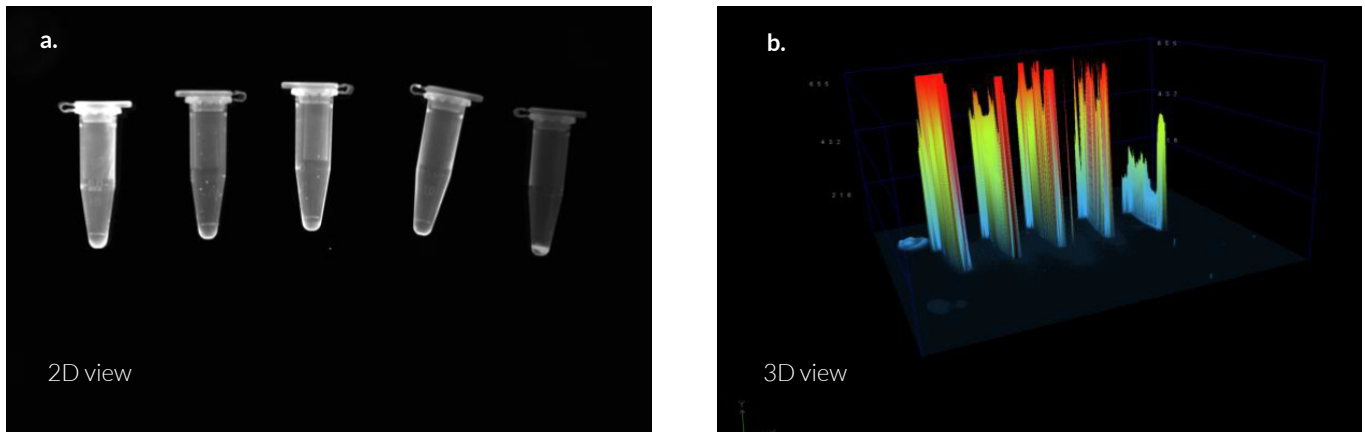


Figure 3. Light diffusion with Spot LED based technology

3a. 2D view of 2ml test tubes with GFP under LED spot at exc. 480nm and em. 550nm **3b.** 3D view of poor light homogeneity in test tubes by grey level comparison (UVITEC Alliance software)

Our Chromascan concept (Fig. 4.) addresses this issue of homogeneity with a sophisticated scan-like concept based on the power of LED excitation. The flexibility of our 12 Chromascan clip-and-play module packs allows you to scan multiple proteins with unparalleled light homogeneity.

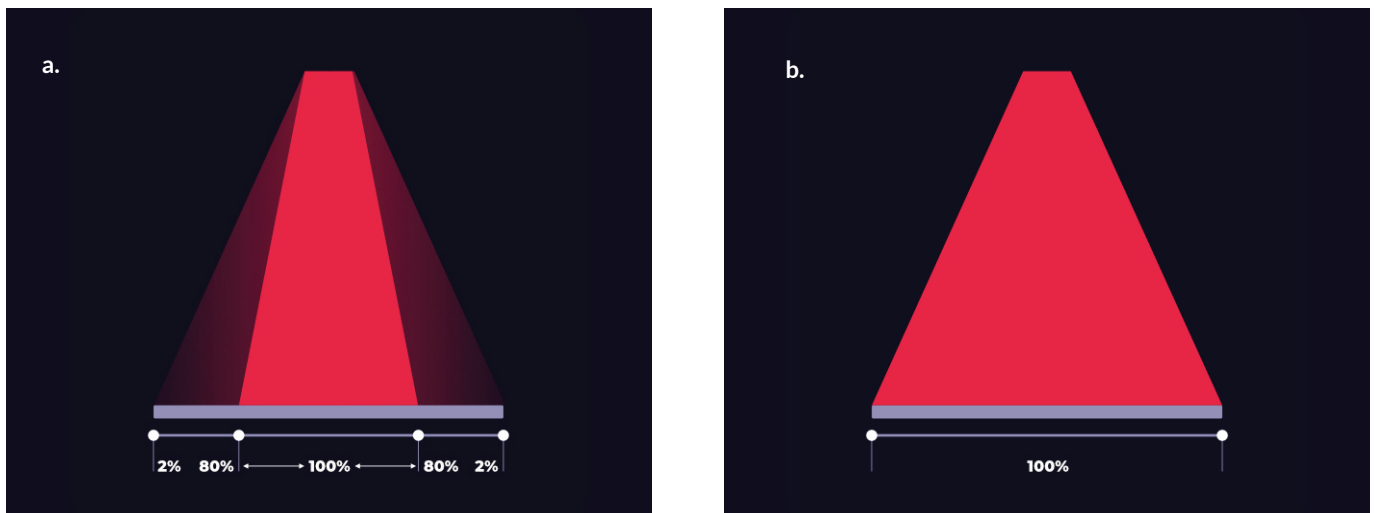


Figure 4. Multiplexing with Chromascan

4a. Non-homogeneity of light providing in common spot-LED technology **4b.** Chromascan, new light source increasing homogeneity of light

■ Reliable and customizable multiplexing

Why limit yourself to 5 channels of excitation when you can use up to 8 Chromascan illumination modules in your imaging system ?

To obtain the best possible results, the source of excitation needs to be carefully considered. Alternatively, the more choice you have the greater the number of dyes you will be able to answer. Today, blue and green dyes are more commonly used

due to their versatility and cost-efficiency. However, autofluorescence is a common issue associated to dyes within these wavelengths resulting in significant sample interference. To reduce this variable, excitation wavelengths above 600nm (Red, NIR and IR) have started to generate significant interest as background noise is reduced and when working with living cells, as cellular damage is shown to be lowered **(2, 3)**.

■ 12 combinations to match your needs

To answer the various numbers of epifluorescent dyes that may be used in a laboratory, we have designed 12 different packs of excitation and emission sources for you to easily insert into your system - you won't even need a technician to do this **(Fig.5.)**. This allows you to gradually expand, test and interchange your epifluorescence studies whenever you wish, through a Clip-and-Play concept.

Chromascan Modules	Application Dyes
Chromascan UV365 pack module EX365nm-EM590nm	Qdot 565, Qdot 655, Qdot 705, TLC plates, microplate, Alexa 350, DAPI
Chromascan Light Blue pack module EX440nm-EM500nm	DAPI, CFP, Cerulean, Alexa Fluor 405, Cascade Blue, Pacific Blue, DyLight 405, Atto 425
Chromascan Blue pack module EX480nm-EM550nm	FITC, Alexa Fluor 488, GFP, SYTOX Green, Fluorescein, Cy2, Sypro Ruby, DyLight488
Chromascan Deep Blue pack module EX480nm-EM600nm	YFP, eYFP, Venus, Alexa Fluor 514, FITC, mCitrine
Chromascan Green pack module EX540nm-EM600nm	Rhodamine, Alexa Fluor 532, Alexa Fluor 555, Cy3, PE, TRITC, ProQdiamond, DyLight 549, mRuby3
Chromascan Deep Green pack module EX540nm-EM650nm	Cy3.5, Atto 565, Rhodamine 6G
Chromascan Orange pack module EX580nm-EM650nm	DsRed, mCherry, Cy3.5, Alexa Fluor 568, Texas Red, Atto565, Atto594, Alexa594, mStrawberry, mKate2
Chromascan Red pack module EX640nm-EM700nm	Alexa Fluor 647, Alexa Fluor 660, Cy5, APC, Atto633, Atto 647N, DyLight 633, DyLight 650
Chromascan NIR pack module EX680nm-EM750nm	Alexa Fluor 680, Alexa Fluor 700, Cy5.5, IRDye 680RD, Atto 680, Atto 700, APC-Cy7, DyLight 680
Chromascan LIGHT IR pack module EX740nm-EM750nm	Alexa Fluor 750, Cy7, IRDye 800CW, VivoTag-S 750, DyLight 755
Chromascan IR pack module EX740nm-EM800nm	Cy7, IRDye 800RS, ZW800-1, Atto 740, HiLyte Fluor750
Chromascan FAR IR pack module EX780nm-EM850nm	Alexa Fluor 790, Cy7.5, IRDye 800CW, VivoTag-S 800, DyLight 800

Figure 5. Chromascan modules packs

*Spectral curves on demand

Wide choice of preset Chromascan modules packs for all your applications



Each of our high-end modules comes with the appropriate confocal emission filter (**Fig.6.**) to avoid crosstalk and be certain to observe your desired and specific fluorophore (**Fig.7.**).

Figure 6. Excitation and emission sources

Each chromascan module pack includes a light excitation source and an emission filter

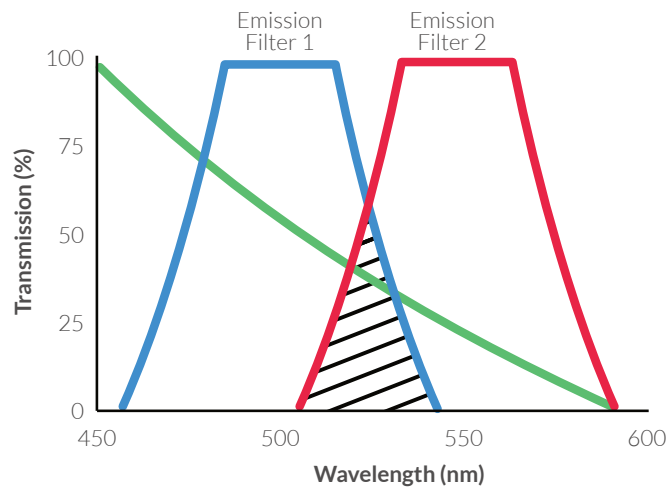


Figure 7. Crosstalk and bleedthrough

Crosstalk occurs when emission filter (blue) cross-overs in the neighbouring emission filter (red) causing signal (green) to be accepted by both filters (4)

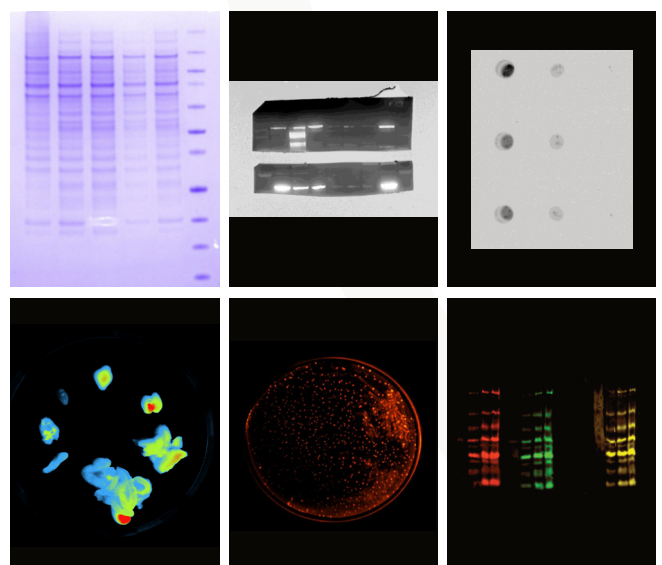
With its large choice of module packs, Chromascan ensures a more reliable and high performing multiplexing experiment with limited crosstalk and heightened accuracy.

■ Endless applications

The Alliance IRIS, equipped with Chromascan technology, represents a significant advancement in the field of molecular biology imaging. By providing uniform illumination, accurate quantification and superior multiplexing capabilities, Chromascan empowers researchers to achieve high-quality, reproducible results.

With Alliance IRIS, your application coverage is infinite:

- Chemiluminescence (**5, 6, 7**),
- Epifluorescence (**8, 9, 10**),
- RNA-DNA Gels,
- Coomassie gels (**11**),
- Elisa Plate,
- Bioluminescence,
- Colony counting, and many more...



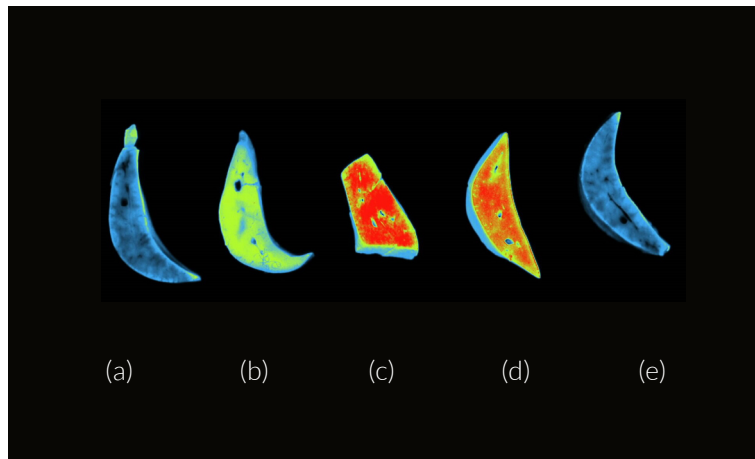


Figure 8. Monitoring nanoparticle distribution in rat organs

Rhodamine staining with Alliance Chromapure exc. 580nm and em. 650nm. (a) 0 day control group, (b) 2 days, (c) 1 week, (d) 1 month and (e) 2 months. Results indicated after 2 months that the liver has the same signal intensity to the control group indicating that nanoparticle distribution took 2 months to clear liver. Samples from Dr. Chiara Castellani, University of Padova, Italy

■ References

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- (2) Effect of Metalation on Porphyrin-Based Bifunctional Agents in Tumor Imaging and Photodynamic Therapy / Patel *et al.* 2016 (*Bioconjugate Chemistry*)
- (3) In vivo near-infrared fluorescence imaging / Frangioni. 2003 (*Current opinion in chemical biology*)
- (4) Fluorescence Resonance Energy Transfer (FRET) / Herman *et al.* (*Olympus Life Science*)
- (5) Neurodevelopment disorder in Zebra fish/ Fasano *et al.* 2022 (*Nature*)
- (6) Diabetic wound models / Huang *et al.* 2024 (*Nature*)
- (7) Breast Cancer Cells / BScumaci *et al.* 2020 (*Cells*)
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- (9) Muscle protein synthesis/ Patel *et al.* 2019 (*Plos One*)
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WITH
IRIS
MAKE IT VISIBLE