

News Flash from Our Cell Biology Portfolio

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Presentation Outline

1

GloMax Galaxy

2

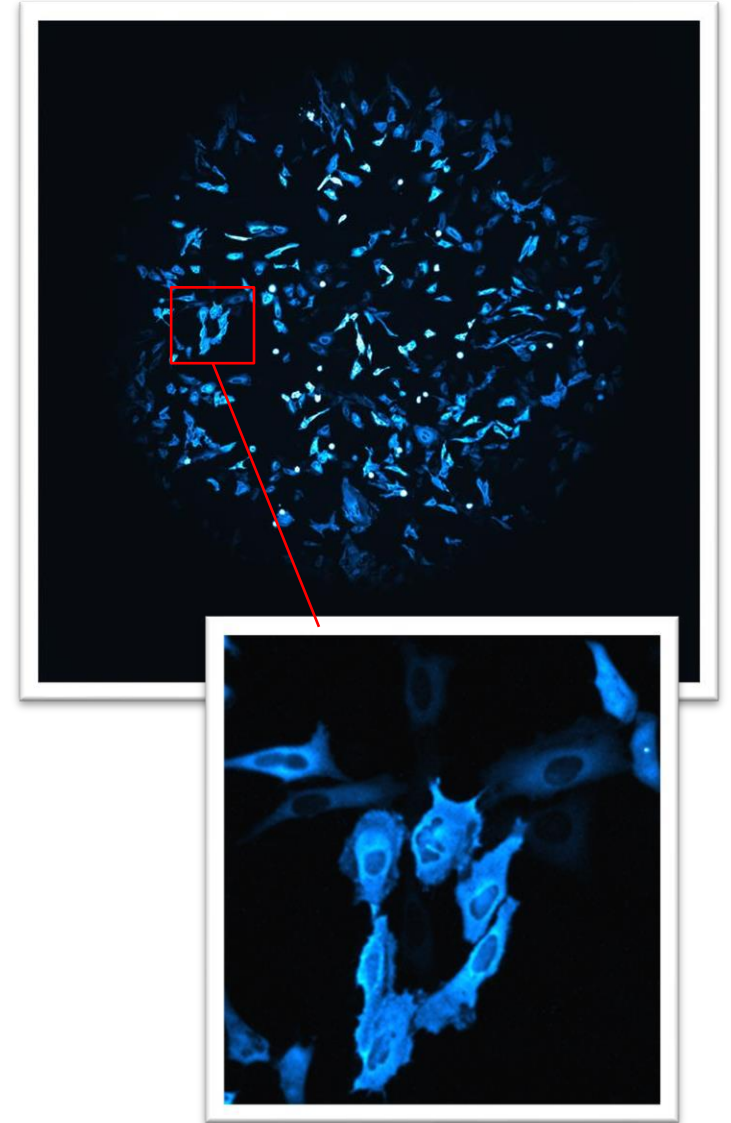
Combining Impedance with Bioluminescence

3

News Flash from Cell Biology Portfolio

Bioluminescence Imaging

- ⌘ Seeing is believing – key advantages of imaging
 - ⌘ Identification of rare events
 - ⌘ Assay validation
 - ⌘ Responders vs non-responders
 - ⌘ Analysis of mixed cell populations
 - ⌘ Spatial localization of the signals (e.g. membrane protein translocation)
- ⌘ Bioluminescence allows viewing **dynamic events** and localization of targets with several benefits:
 - ⌘ No phototoxicity, no photobleaching
 - ⌘ Very low background signal
 - ⌘ Vast functional toolbox for studying different aspects of cell biology
- ⌘ Challenges:
 - ⌘ Low photon flux requires long exposure (seconds to minutes)
 - ⌘ Expensive instrumentation \$\$\$



GloMax Galaxy Bioluminescent Imager

- ✂ Use NanoLuc® technologies to study rare events and analysis of mixed cell populations
- ✂ Study protein dynamics and cellular physiology
- ✂ Living & fixed cells & tissues
- ✂ Ideal for assay development

LUMINESCENCE

Protein dynamics
and localization

FLUORESCENCE

Cellular reference
markers

BRIGHTFIELD

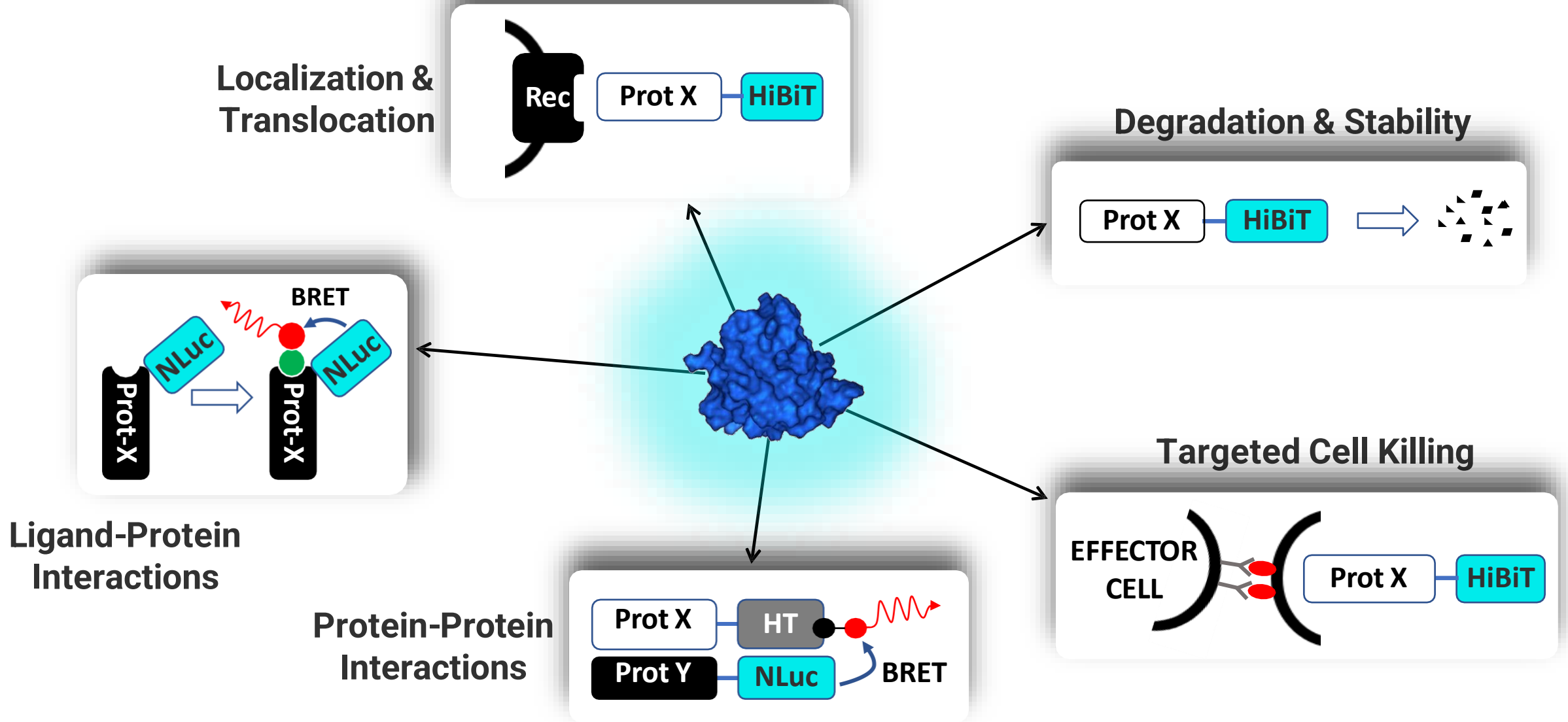
Morphology

Affordable, Easy to use, Low-throughput



- ✂ Includes PC and monitor
- ✂ Compatible with slides, microchambers, dishes, and plates
- ✂ Motor-driven focusing and alignment
- ✂ 20X objective lens (10X overall magnification)
- ✂ Accessory: Environment Chamber (temperature, humidity, gas)

Protein Reporter Applications of NanoLuc Technologies

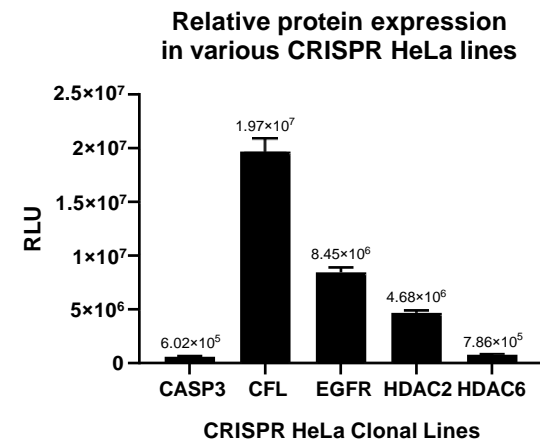


Imaging Low Abundance Endogeneous Proteins

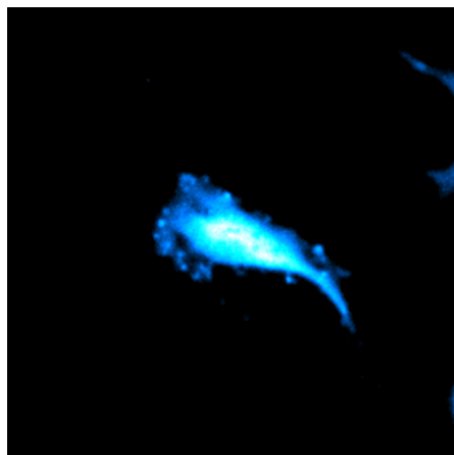
HiBiT inserted to genomic locus via CRISPR/Cas9 in HeLa cells

LgBiT expressed ectopically

Binary Complementation of NanoBiT® Enzyme

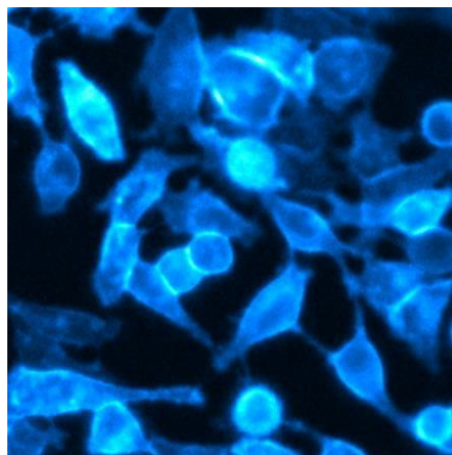


Cofilin



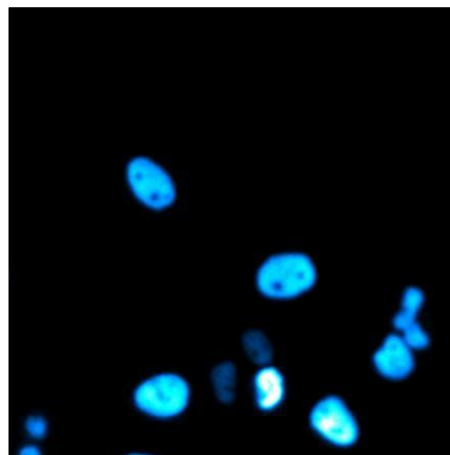
1-minute exposure

EGFR



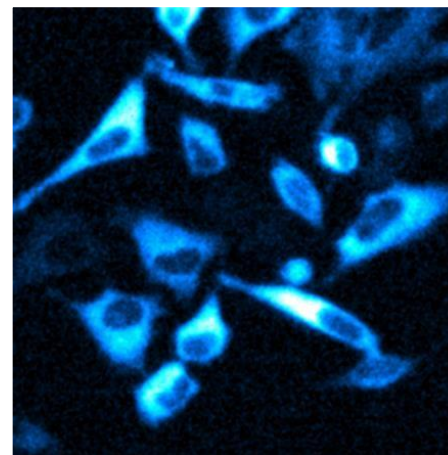
1-minute exposure

HDAC2



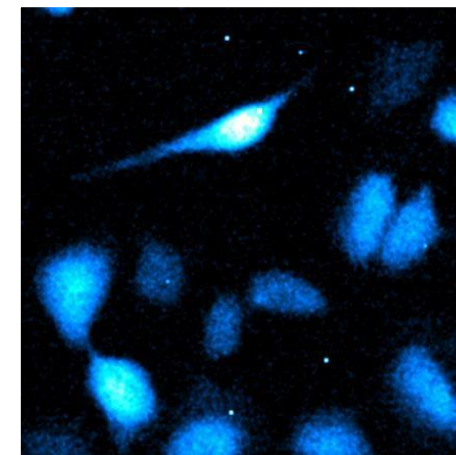
1-minute exposure

HDAC6



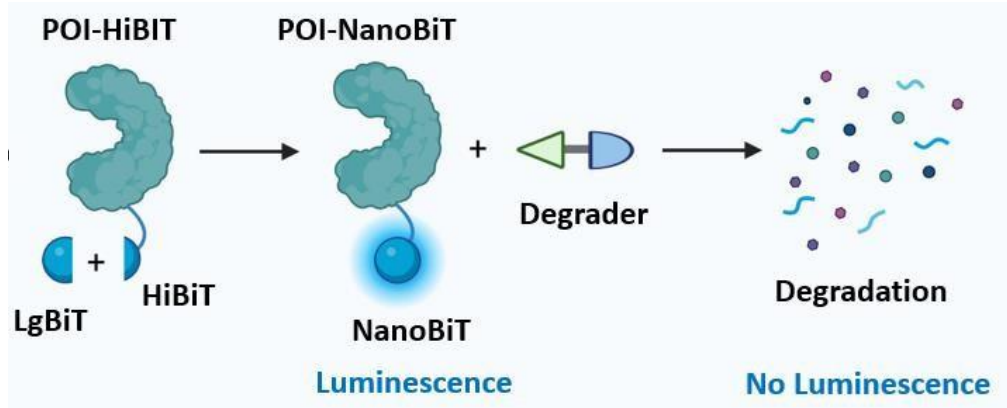
Low Expression
~3-minute exposure

CASP3

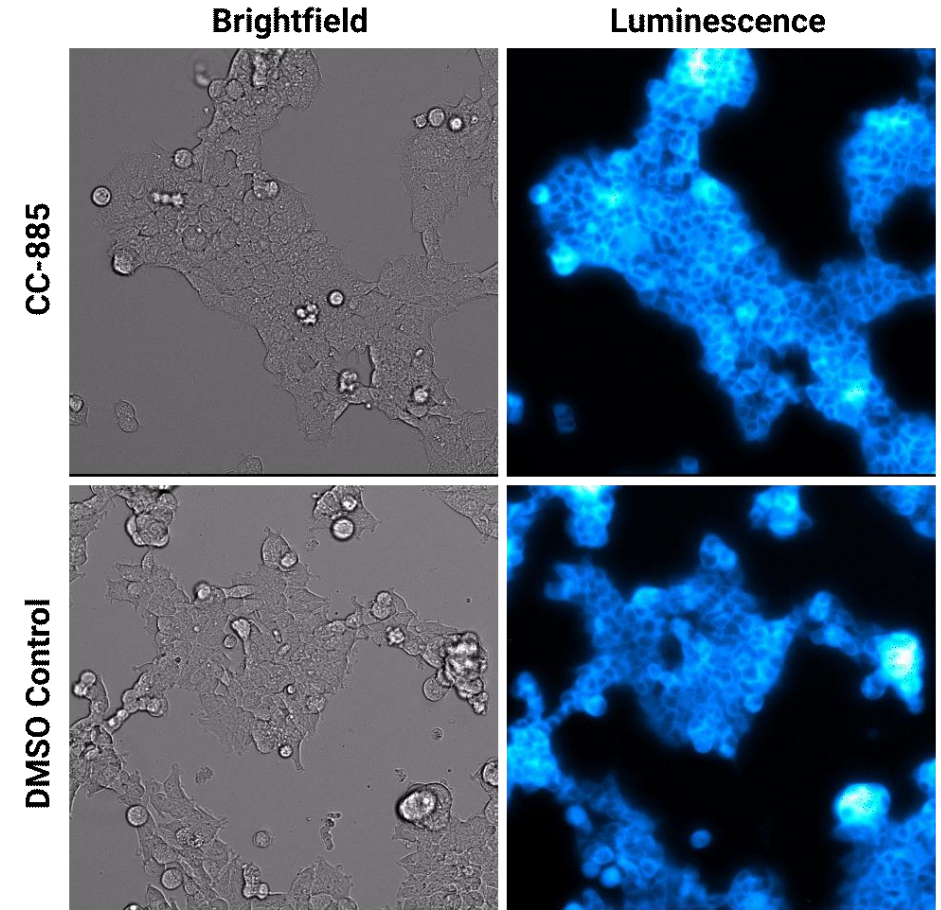


Very Low expression
~5-minute exposure

Targeted Protein Degradation of Endogenous GSPT1



- ✂ HEK293 cells expressing endogenous HiBiT-tagged GSPT1 and stably expressing LgBiT were treated with CC-885 degrader or DMSO control treatment.
- ✂ Assayed with Nano-Glo® Vivazine Live Cell Substrate and imaged over 5 hours in stagetop incubator.
- ✂ Molecular-glue degrader, CC-885, facilitates targeted protein degradation of GSPT1, and acts as an anti- tumor agent.

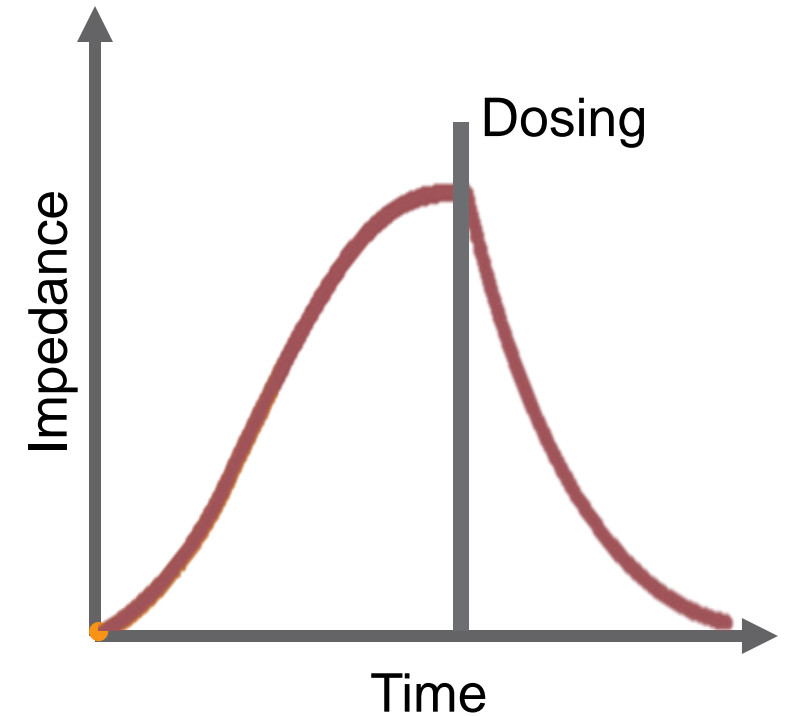
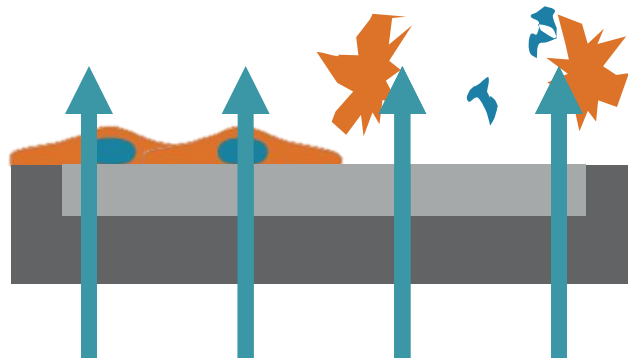


Presentation Outline

- 1 GloMax Galaxy
- 2 Combining Impedance with Bioluminescence**
- 3 News Flash from Cell Biology Portfolio

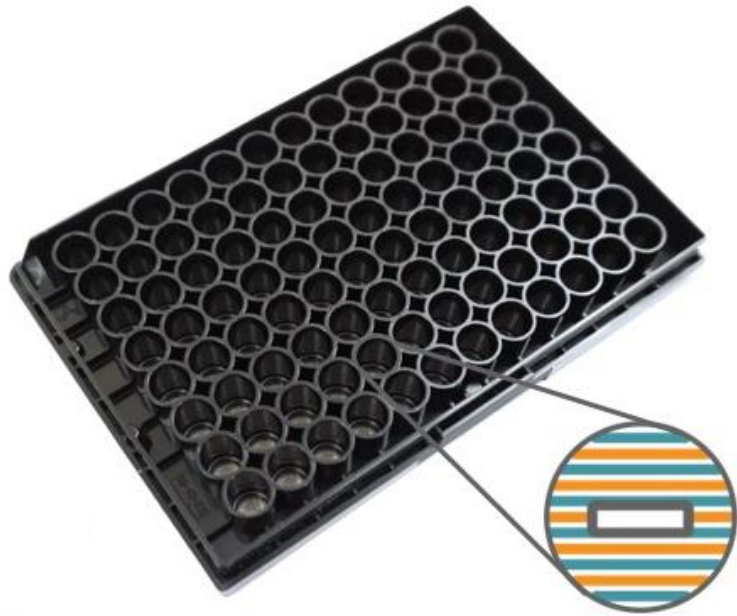
Impedance Assays Principle

- ✂ Measures how easily signal passes the electrode-cell interface
- ✂ Resistance increases as coverage and attachment increases
- ✂ Can detect:
 - ✂ Proliferation
 - ✂ Viability
 - ✂ Cell-cell coupling strength (barrier function)
 - ✂ Migration
 - ✂ Cell signaling

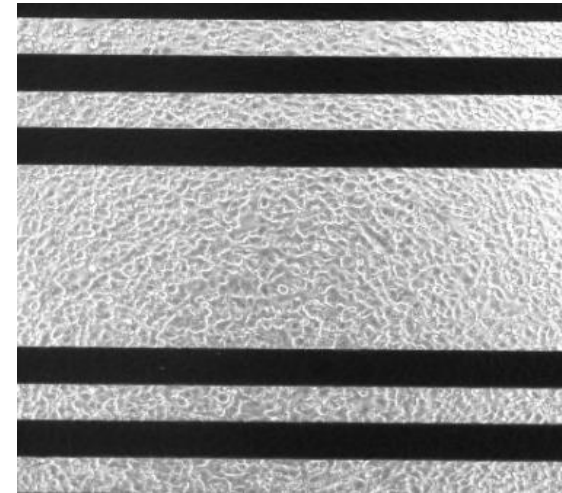


Impedance Assays Principle

- ✂ CytoView-Z plates have a recording electrode embedded in the culture surface of each well.



- ✂ Culture cells in the plate
- ✂ Viewing window in 96-well plate allows cell visualization and assay multiplexing



Impedance Assays vs. Traditional Assays

✂ Captures all stages of an experiment

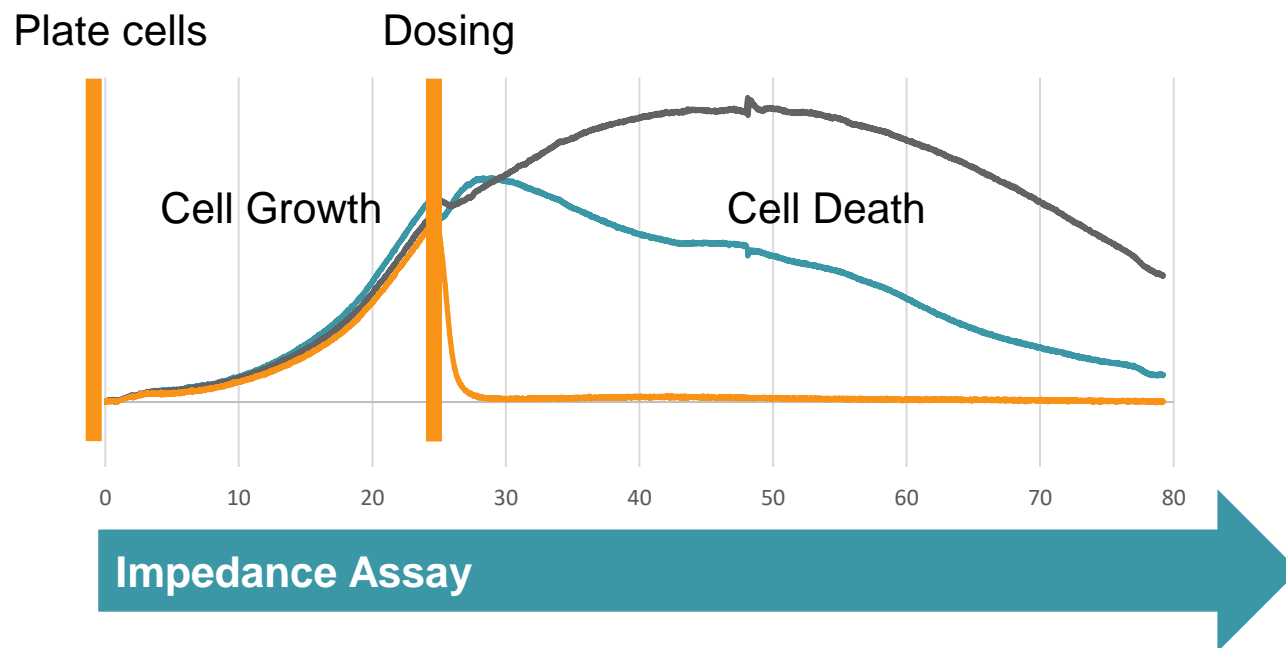
- ✂ Cell growth and death
- ✂ Acute or chronic treatments
- ✂ TEER measurements

✂ Hands-free data collection

- ✂ Plate cells, add treatments, done

✂ Label-free

- ✂ Measurement doesn't impact biology
- ✂ No optimization of labels, dyes, or incubation times required
- ✂ Multiplex with bioluminescent and fluorescent cell-based assays



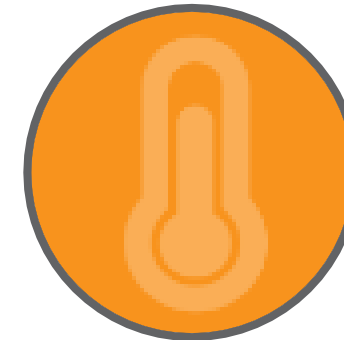
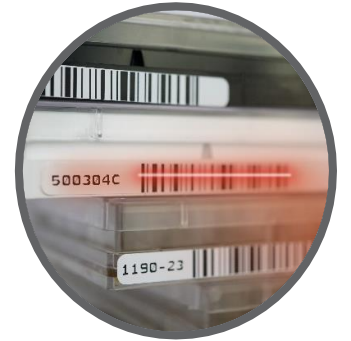
Maestro Z Advantages

- ✂ One-button setup with barcode tracked plates
- ✂ Automated CO₂ and temperature control
- ✂ Automatic event tracking records door movements
- ✂ Multiplex measurements in the plates
- ✂ No computer needed while measuring
 - ✂ Only required for initiation and analysis
- ✂ Small footprint
- ✂ Mobile app for remote monitoring



**Push-button
Acquisition**

**Barcode Tracking
& Audit Trail**

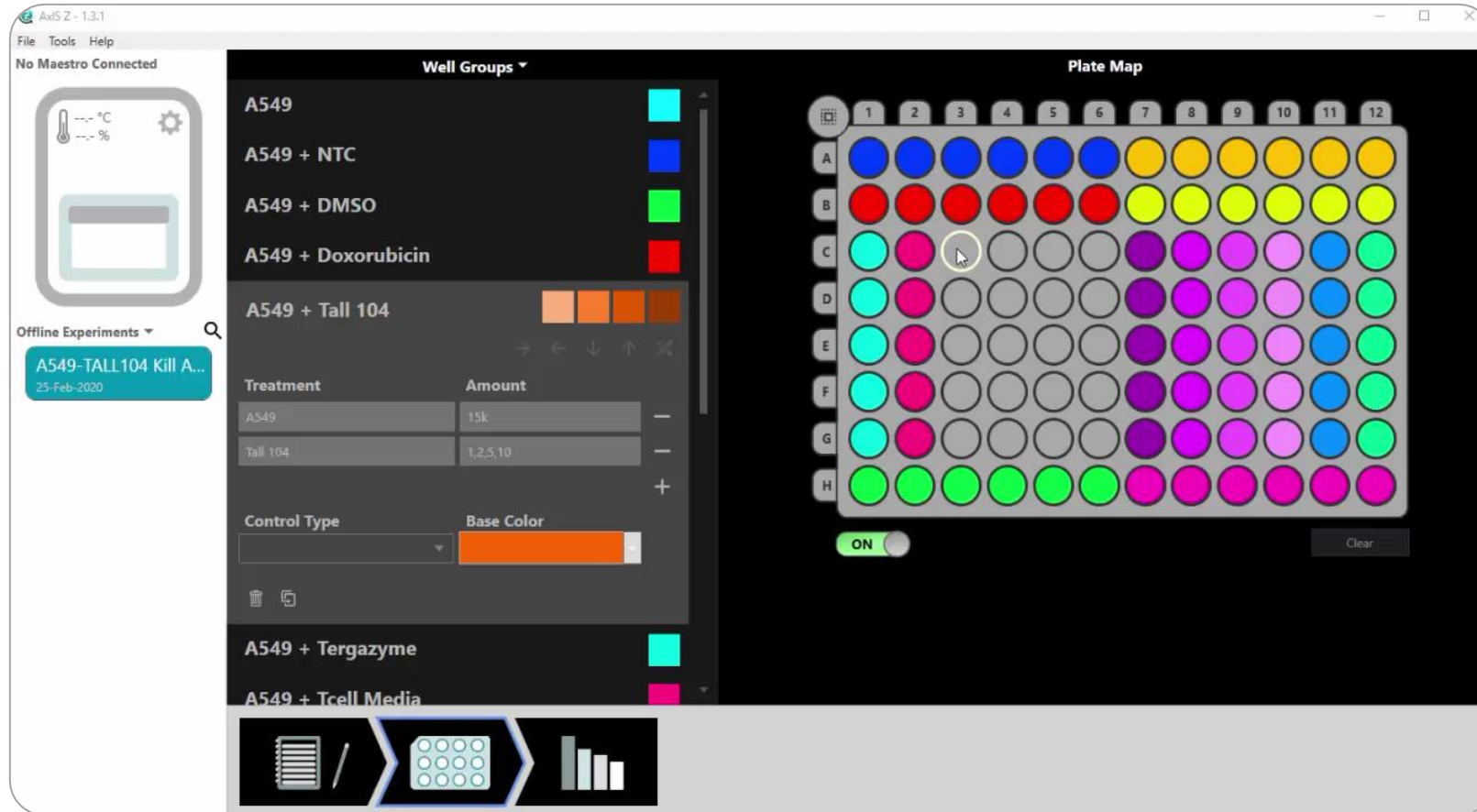


**Automated
Environmental
Control**



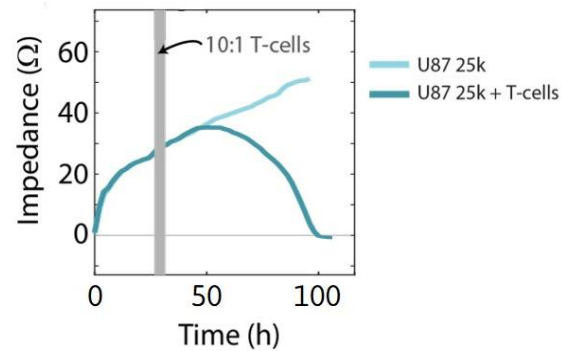
Label-free, continuous monitoring of cell behavior

Quantitatively track cell proliferation, viability, and cytotoxicity

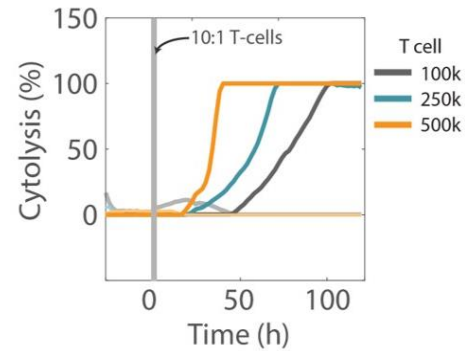


The only impedance assay that can be performed with a single-click

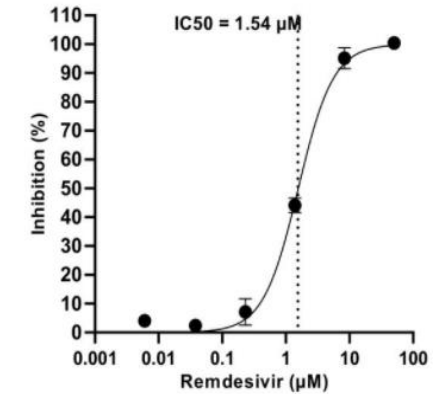
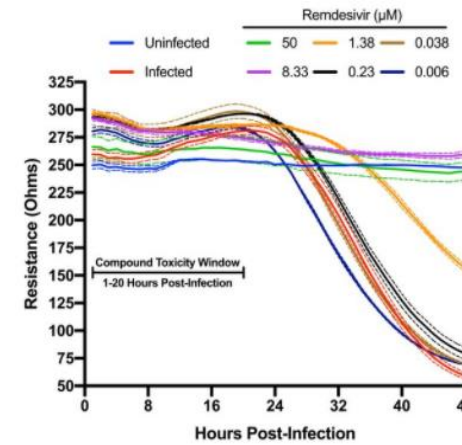
Impedance Measurement Applications



Immune cell kill assay

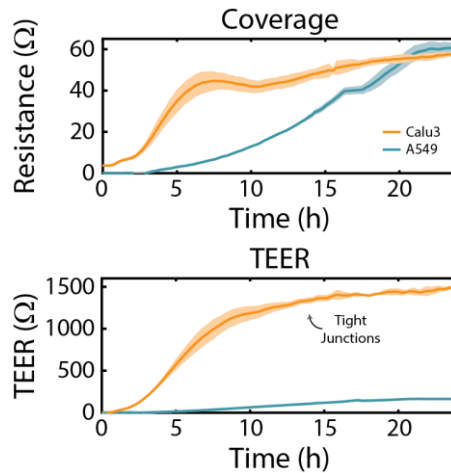


Kill curve

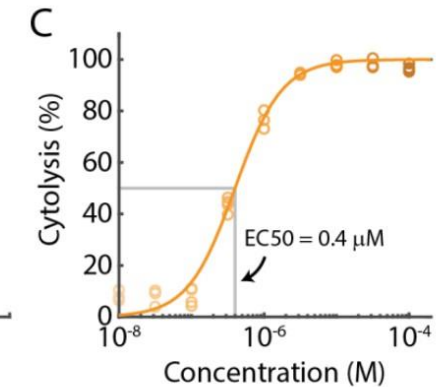
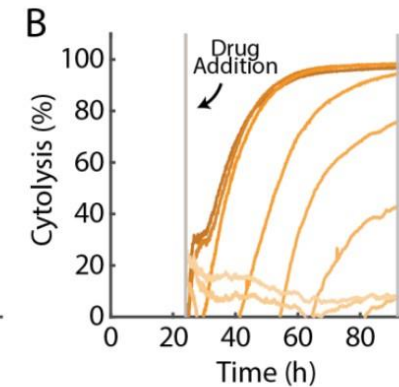
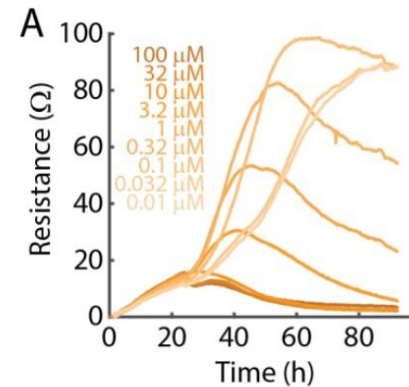


✂ CAR-T therapy development

✂ Viral cytopathic effects



✂ Transepithelial electrical resistance (TEER) changes

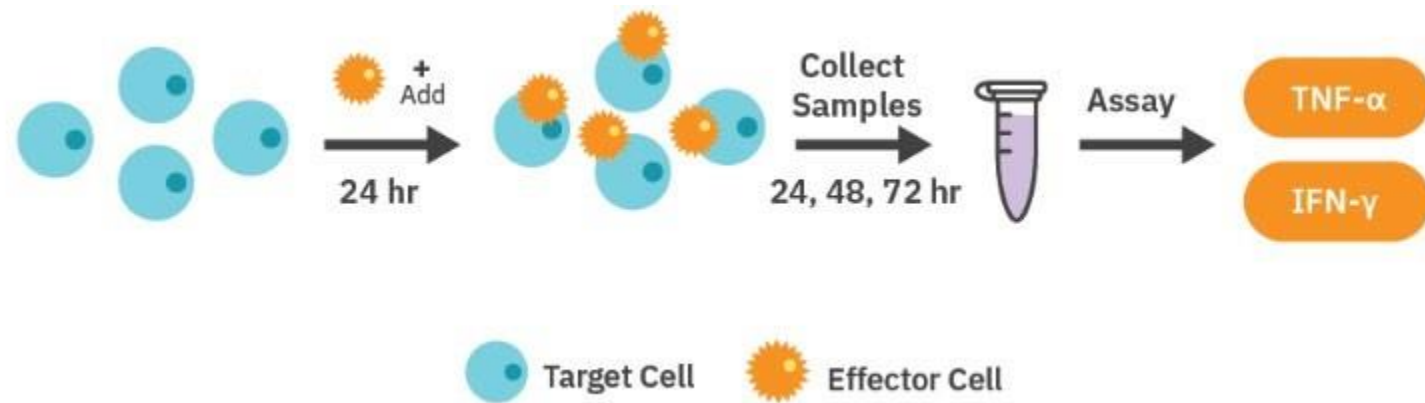


SKOV3 cells treated with 9 different concentrations of doxorubicin

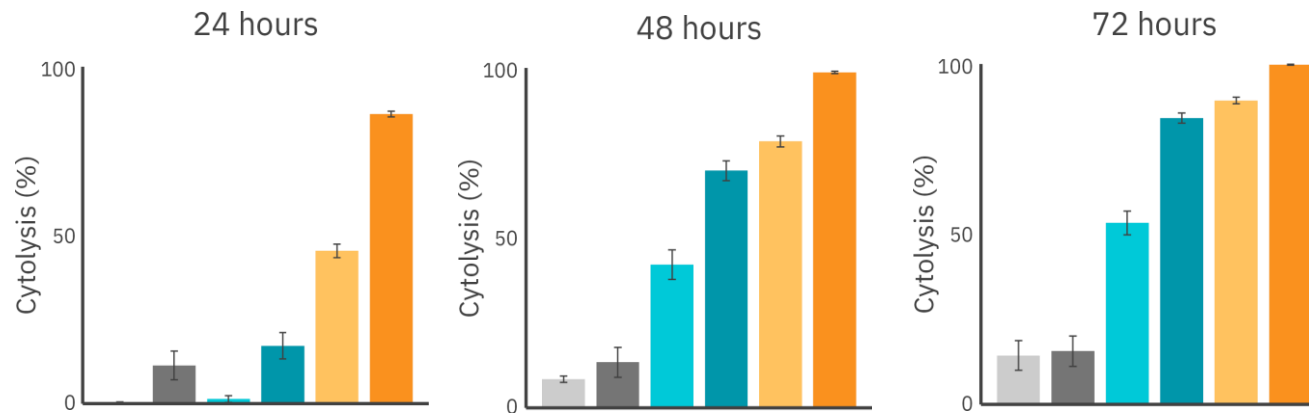
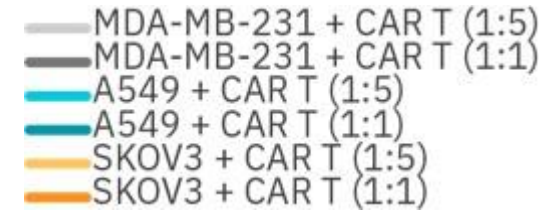
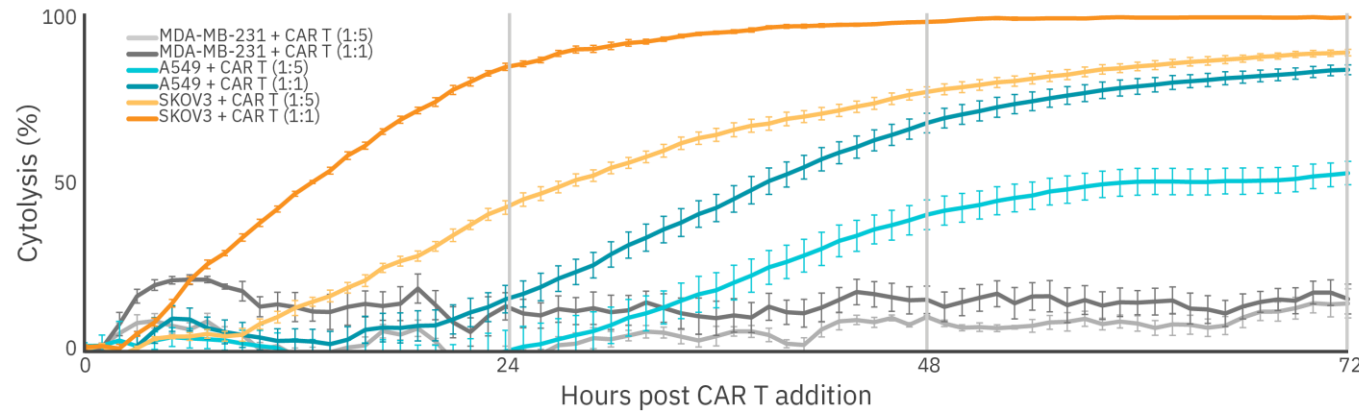
✂ Dose-response analysis

Combination of Impedance with Bioluminescent for CAR-T development

- ⌘ Does tumor antigen density impact CAR-T Cell performance?
- ⌘ HER2 CAR-T cells were cocultured with:
 - ⌘ SKOV3 (high HER2 expression)
 - ⌘ A549 (low HER2 expression)
 - ⌘ MDA-MB-231 (no HER2 expression)
- ⌘ CAR-T cell killing monitored by impedance, TNF- α & IFN- γ detected by Lumit Immunoassays

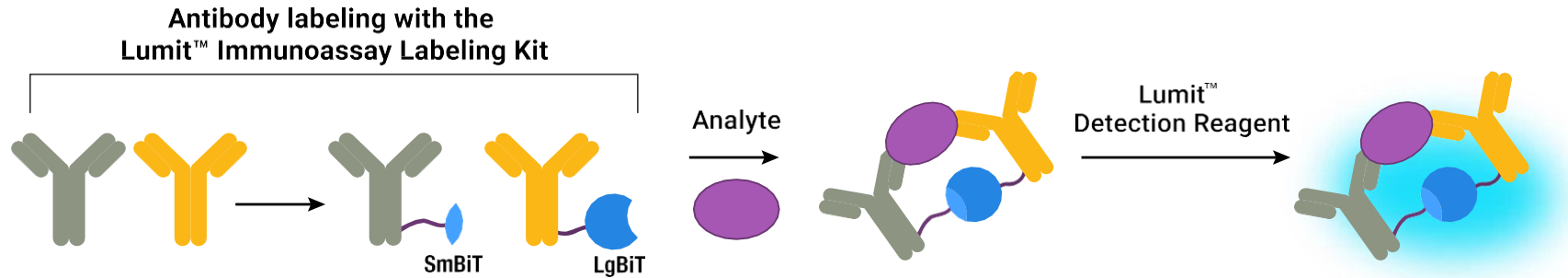


Combining Impedance with Bioluminescent Assays for CAR-T development

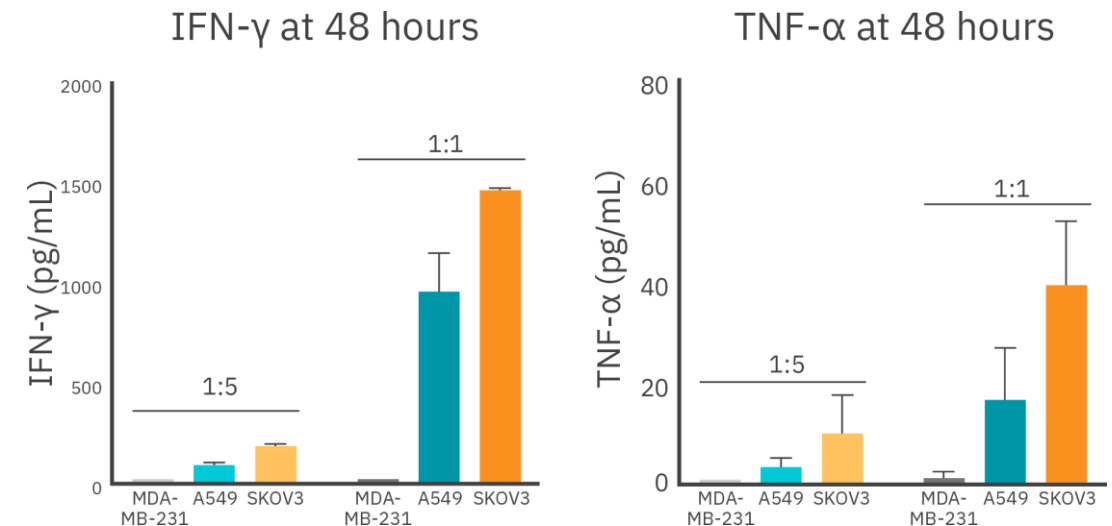


- CAR-T cell killing correlated with antigen expression levels
- Observed differences in cell killing change over time
- MDA-MB-231 cells showed 20% cytotoxicity due to nonspecific killing

Combining Impedance with Bioluminescent Assays for CAR-T development



- ✗ CAR T cells co-cultured with SKOV3 (high HER2) released 41.6% more IFN- γ compared to A549 (low HER2)
- ✗ CAR T cells co-cultured with SKOV3 released 80.5% more TNF- α compared to A549
- ✗ CAR T cells co-cultured with MDA-MB-231 (no HER2) did not release detectable TNF- α or IFN- γ



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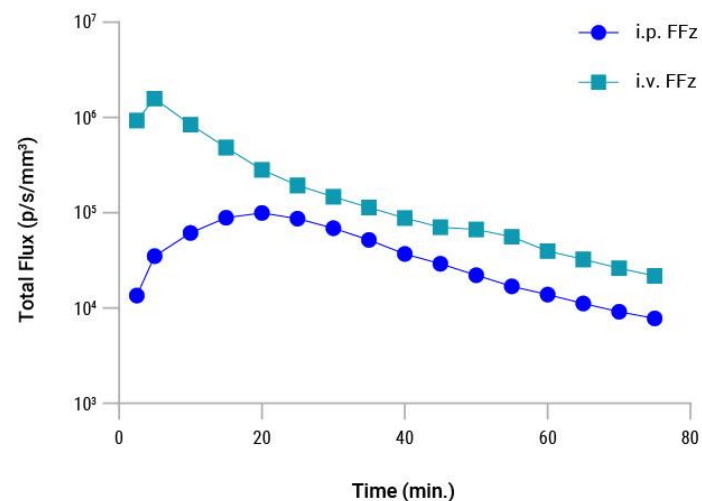
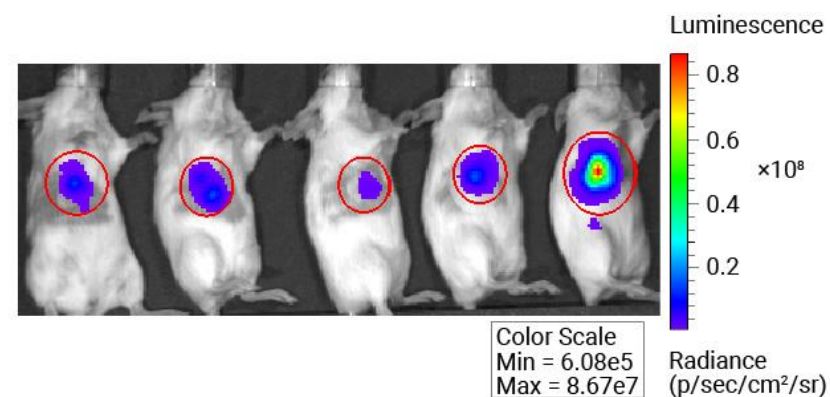
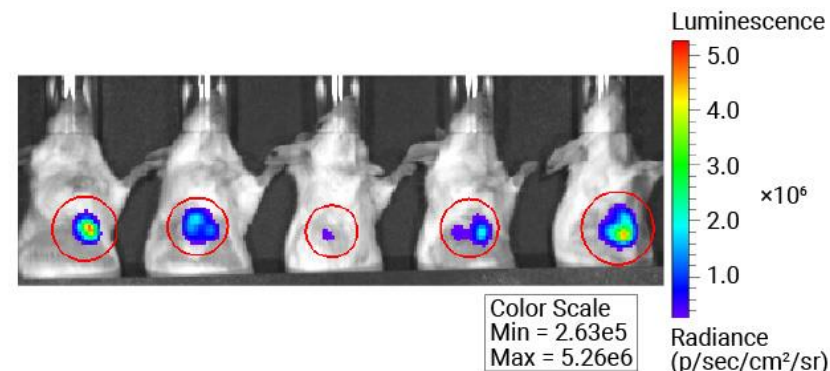
Combining Impedance with Bioluminescence

3

News Flash from Cell Biology Portfolio

Imaging NanoLuc In Vivo

- ✂ Nano-Glo Fluorofurimazine has increased aqueous solubility and allows increased substrate delivery
- ✂ Brighter in vivo signal and increased signal stability
- ✂ Greater flexibility in delivery options – intraperitoneal vs intravenous injection
- ✂ NanoLuc's small size makes it easy to pack into viral genomes and track virus tissue penetration for gene therapy or infectious disease applications



High Quality Cell Culture Media and Sera



- German company established in 2013
- Specialises on the production of high quality sera and cell culture media and reagents
- Possibility of custom manufacturing from 20 liters



Sera

- FBS
- Other bovine and animal sera
- Human sera



Cell Culture Media

- Classic liquid media
- Classic powdered media
- Special culture media
- Cryopreservation



Cell Culture reagents

- Supplements and additives
- Antibiotics
- BSA
- Trypsin
- Cell separation



Balanced Salt Solutions

- Liquid buffers
- Powdered buffers



Diagnostics

- Virology media
- Cytogenetics

High Quality Sera for Cell Culture



FBS Standard	FBS Advanced	FBS Xtra
Natural FBS	Slightly reduced raw FBS content	Reduced raw serum content
Low endotoxin	Cost-efficient with defined additives	Chemically defined additives
Consistent quality	Minimal lot-to-lot variations	Reduced lot-to-lot variations
Pricing highly dependant on raw FBS price fluctuations	No further batch testing necessary	More sustainable ingredients
Suitable for all cell types	Best for tumor cells & less demanding cell types	Best for tumor cells & fast-growing cell types

High Quality Sera for Cell Culture



Until end of 2024, order FBS Minis for the price of standard 500ml bottles.

Primary Cells, Stem Cells and Media **Lonza**

- Primary cells - over 150 human and animal cell types available
- Clonetics media a growth factors for wide spectrum of primary cells
- Stem cells together with media
- Blood and immune cells from vast collection of donors and sources
 - Specialized X-Vivo™ media



Mycoplasma Testing

- Widespread contamination in a variety of cell culture systems
- Size below 1 μm , hardly visible in optical microscope
- BL MycoAlert™ kit utilizes the Mycoplasma enzymes to convert ADP to ATP
- Converted ATP is consumed by firefly luciferase to produce bioluminescence in case of contamination
- Requires only 100 μl of centrifuged medium for the assay

Lonza

Cell culture + mycoplasma



Cell culture - mycoplasma



Addition of ADP +
MycoAlert™ substrate

ATP

No ATP

Light



Thank you for your attention!

