Multiplexed, live content cellular imaging enabled:
Cell Player™ reagents, assays and IncuCyte Zoom™

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TOOLS, REAGENTS, ASSAYS

IncuCyte Zoom Live Cell Imaging Device resides within a standard cell culture incubator.

Cell culture consumables (e.g. micro-titre plates, T-flasks, petri dishes) are placed within for in situ assays.

Gathers time lapse images from cells – definition phase contrast, green and red fluorescence.

User interchangeable objectives – 4x, 10x, 20x.

Up to 6 x 384-well plates simultaneously.

Simple to use but highly advanced phase and fluorescent image processing software tools.

Targeted GFP & RFP lentiviruses

3rd generation HIV-based, VSVG pseudotyped lentiviral particles encoding cytoplasmic (Cytoplasm) or nuclear restricted (NucLight) GFP or RFP, or a combination of the two (Duolight).

Expression driven off either an EF-1a or CMV promoter with antibiotic resistance cassette for stable cell line generation.

Validated as non-perturbing to cell health across a range of MOIs.

Lentiviral targeted GFP & RFP

<table>
<thead>
<tr>
<th>Catalog Name</th>
<th>Type</th>
<th>Localization</th>
<th>Promoter</th>
<th>Selection</th>
</tr>
</thead>
<tbody>
<tr>
<td>4475</td>
<td>Lentivirus</td>
<td>Nucleus</td>
<td>EF-1a</td>
<td>Puromycin</td>
</tr>
<tr>
<td>4476</td>
<td>Lentivirus</td>
<td>Cytoplasm</td>
<td>CMV</td>
<td>Puromycin</td>
</tr>
<tr>
<td>TBD</td>
<td>DuoLight (Red/Green)</td>
<td>Lentivirus</td>
<td>Nuc + Cyto</td>
<td>CMV</td>
</tr>
</tbody>
</table>

Nuclear / Cytoplasmic GFP / RFP stable cell lines

Created by transduction of the host cell with the targeted lentiviruses (above).

Typically >95% of cells express the fluorescent protein.

Seeded in standard culture plates.

Seeded at comparable to host cell lines (morphology, growth rates, migration rates).

Stable cell lines expressing targeted GFP & RFP

<table>
<thead>
<tr>
<th>Seeding No.</th>
<th>Type</th>
<th>Transducer Marker</th>
<th>Destination</th>
</tr>
</thead>
<tbody>
<tr>
<td>4475</td>
<td>Lentivirus</td>
<td>NucLight Green</td>
<td>Lentivirus</td>
</tr>
<tr>
<td>4476</td>
<td>Lentivirus</td>
<td>NucLight Red</td>
<td>Lentivirus</td>
</tr>
<tr>
<td>TBD</td>
<td>DuoLight (Red/Green)</td>
<td>NucLight Green &amp; Red</td>
<td>Bifunctional</td>
</tr>
</tbody>
</table>

VALIDATION OF TARGETTED GFP/RFP LENTIVIRAL REAGENTS & CELL LINES

<table>
<thead>
<tr>
<th>A</th>
<th>AS49 cells</th>
<th>B</th>
<th>HUVEC-DuoLight</th>
<th>C</th>
<th>Neuro-2a NucLight Green</th>
</tr>
</thead>
</table>
| Figure 1 | lentilid infection of immortalised and primary cells. Transduction efficiency of NucLight Green at different multiplicities of infection (MOI) (a) in AS49 (A) and HUVEC (B) cells following 24-48h transduction. Image of AS49 cells expressing the NucLight Green. (C) Note the homogenous nuclear restricted GFP label and healthy appearance of the cells.

PHASE/2-COLOUR ASSAY APPLICATIONS

384-well plate view of kinetic cell proliferation in HT1080-NucLight-Green cells in the presence of different concentrations of cytosine arabinoside (A). (B) Overall immunocass. (C) 384-well assay plate view and IC50 curves.

2-COLOR ADVANCED BIOLOGY MODELS

Angiogenesis: Endothelial and stromal cell co-cultures

Cell invasion: HT1080 and MCF-7 cells

Figure 5: (A) Co-culture of HT1080/NucLight-Red and AS49/NucLight-Green at 1:1 ratio seeding. (B) NucLight software image mask independently identifying red and green nuclei. (C) Time-course of cell count.

NeuroTrack™ Kinetic Neurite Outgrowth

Figure 8: (A) Representative image of Neuro-2A cells. (B) Neuro-2A cells with the quantitation mask applied, identifying neurite outgrowth. (C) Time-course of neurite outgrowth and attenuation by the PEC inhibitor 8c-8220.

3200 \times 800

Object count (1/mm²)

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The data contained in this poster represents work designed and conducted by the entire Essen BioScience R&D team.

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