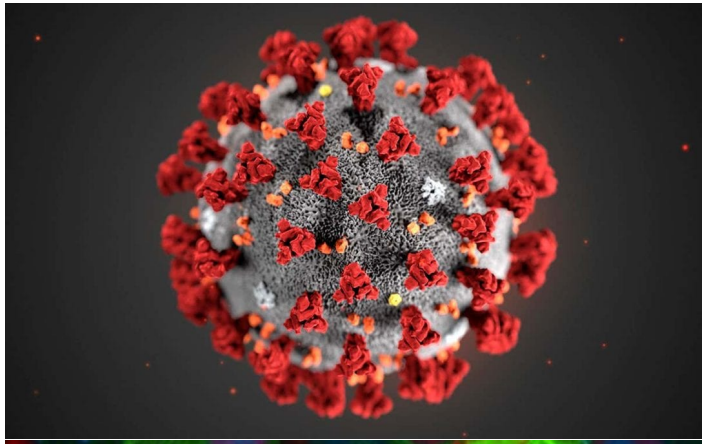
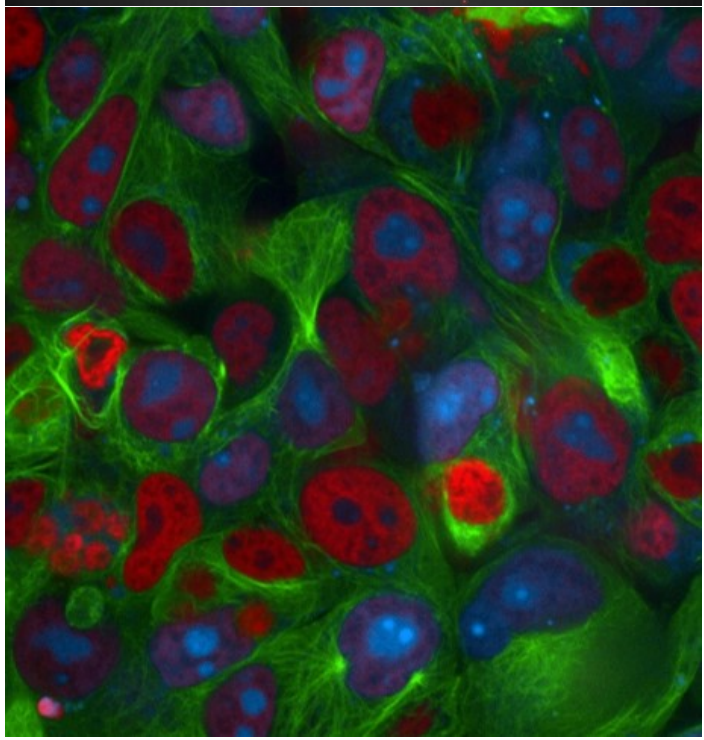
ExpressCells



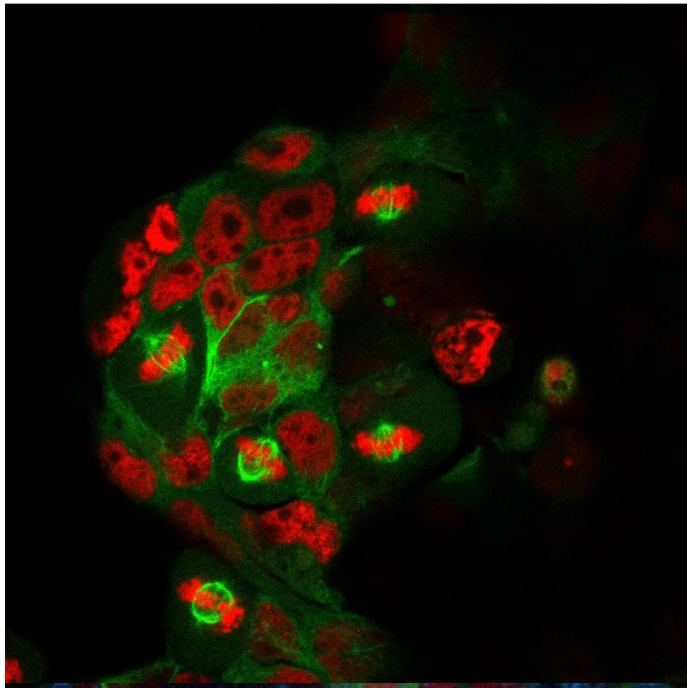


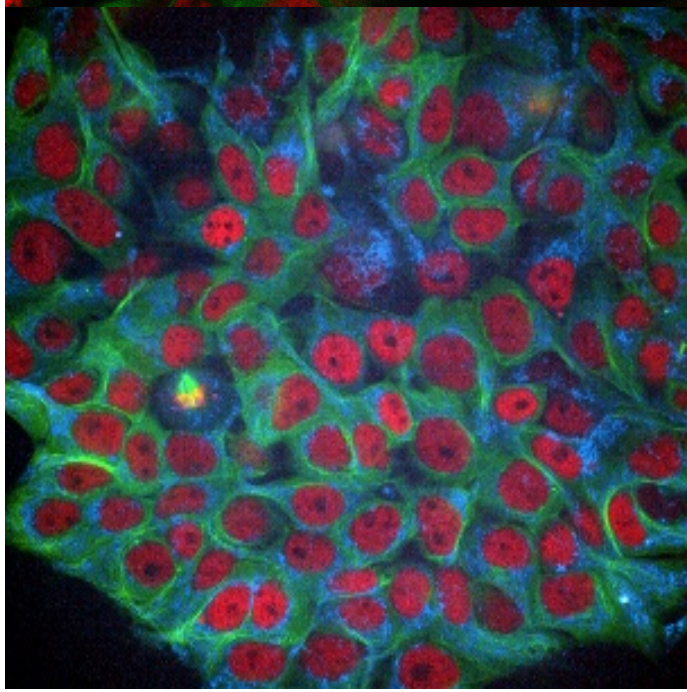
Better knock-ins, better cell lines, better science.

Supporting Research Cellular Biology
(including SARS-CoV-2)

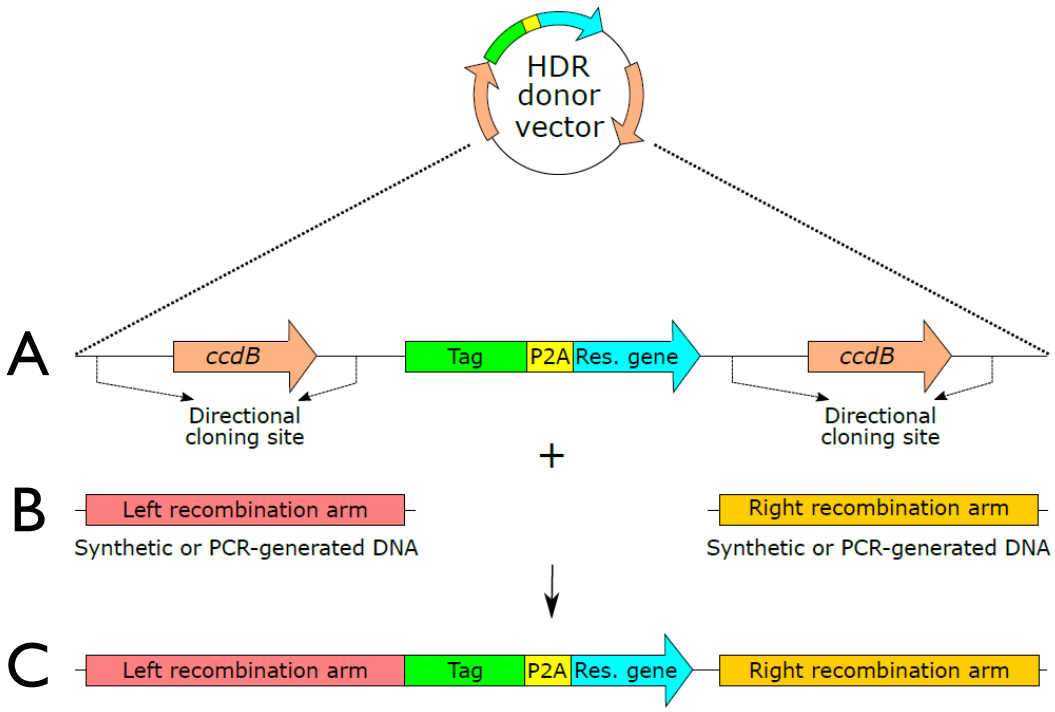
Who We Are

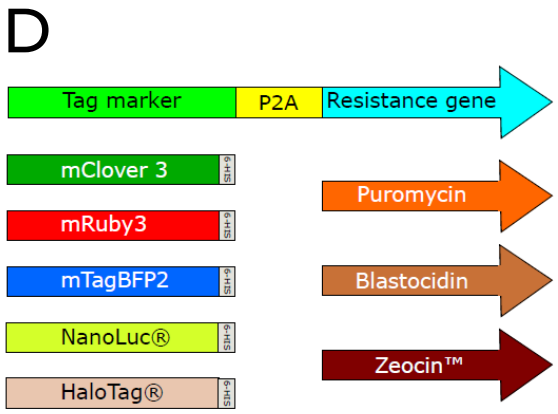
- Genetic engineering company that creates knock-in cell lines for researchers
- Two product options:
  - Catalog: pre-made cell lines that tag specific proteins or support a specific assay
  - Custom: we can knock-in up to three genes in an immortalized mammalian cell line
- Building cell lines that
  - Tag proteins with fluorescent/bioluminescent proteins
  - Overexpress proteins, including those involved in SARS-CoV-2 infection





Our Technology: The FAST-HDR Plasmid Vector System

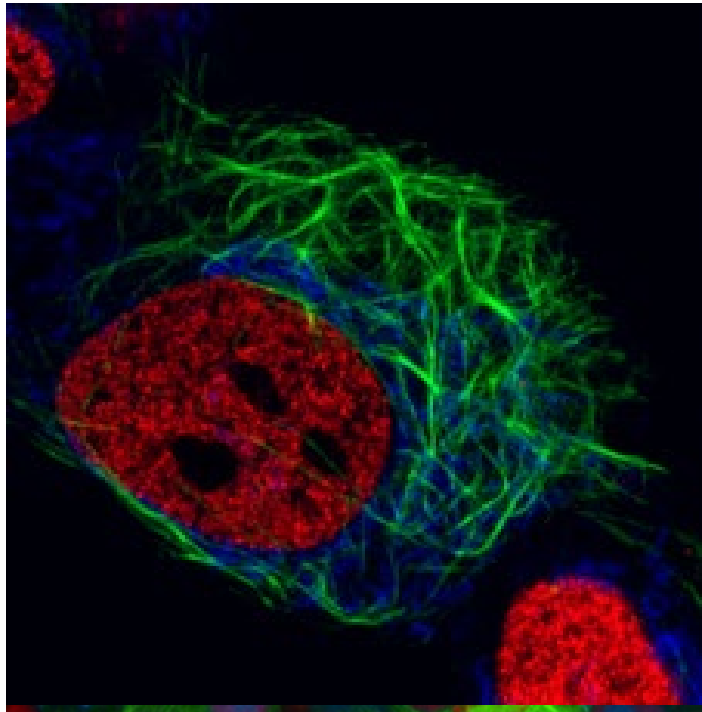


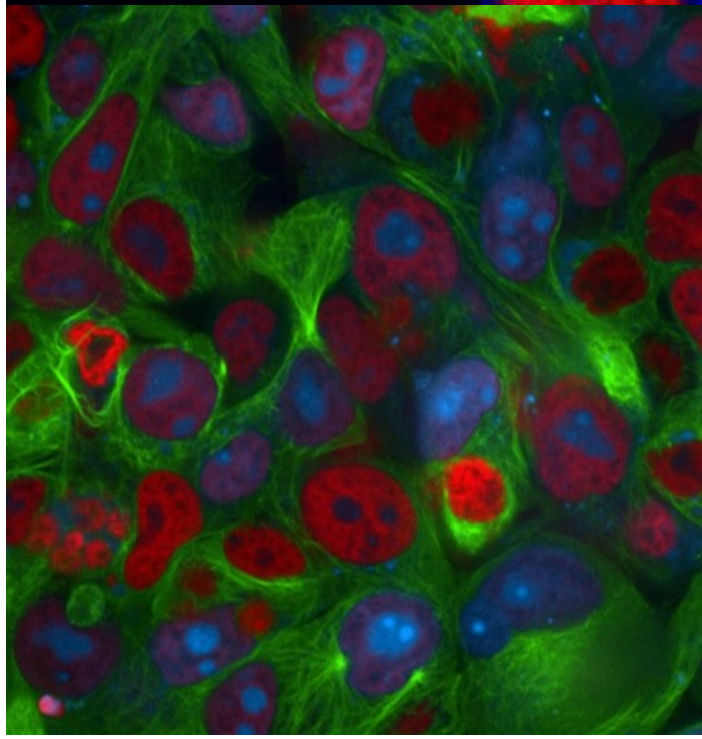


The FAST-HDR Plasmid Vector system, in combination with CRISPR/Cas9, is at the heart of ExpressCells' capabilities, allowing us to deliver custom, knock-in cell lines in 25% the time required with current technologies. We can deliver a cell culture with single knock-in in as few as 30 days and with three in as few as three months.

Figures A – D illustrate construction of the FAST-HDR plasmid vector for C-terminal labeling. In Figure A, a backbone vector is first digested with restriction enzymes excising two *ccdB* cassettes. In Figure B, synthetic recombination arms are mixed and cloned with the digested backbone vector. Figure C depicts the HDR vector product. Figure D illustrates different combinations of reporter tag and antibiotic resistance genes incorporated in backbone vectors.

Cell Catalog





Currently Available Catalog Tagged Human Cell Lines

	Cell Line	Gene or Protein/Tag #1	Gene or Protein/Tag #2	Gene or Protein/Tag #3	Description
EXP-001	HeLa	Ezrin/mRuby3	--	--	Tagged Plasma Membrane
EXP-003	HeLa	TOM20/mRuby3	--	--	Tagged Mitochondria
EXP-004	HeLa	B-Tubulin/ mClover3	--	--	Tagged Cytoskeleton
EXP-005	HEK293T	Histone 3.3/ mRuby3	--	--	Tagged Nucleus
EXP-006	HEK293T	B-Tubulin/ mClover3	--	--	Tagged Cytoskeleton
EXP-007	HEK293T	PARP1/mClover3	--	--	Tagged Nucleus
EXP-008	HeLa	Histone 1/mTagBFP2	TUBB/mClover3	SQSTM1/mRuby3	Autophagy model
EXP-009	HEK293T	B-Tubulin/mClover3	Histone 3.3/mRuby3	ATP5B/mTagBF2	Toxicity Model

- Products in stock
- 3-14 day delivery
- Orders accepted at info@xpresscells.com and https://xpresscells.com/cell-catalog/

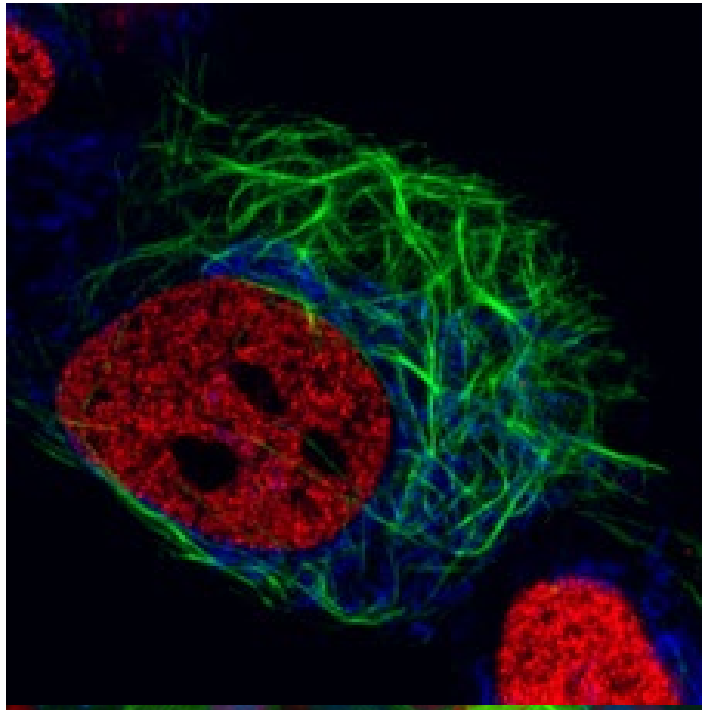
Catalog Human Cell Lines that Overexpress Key Proteins

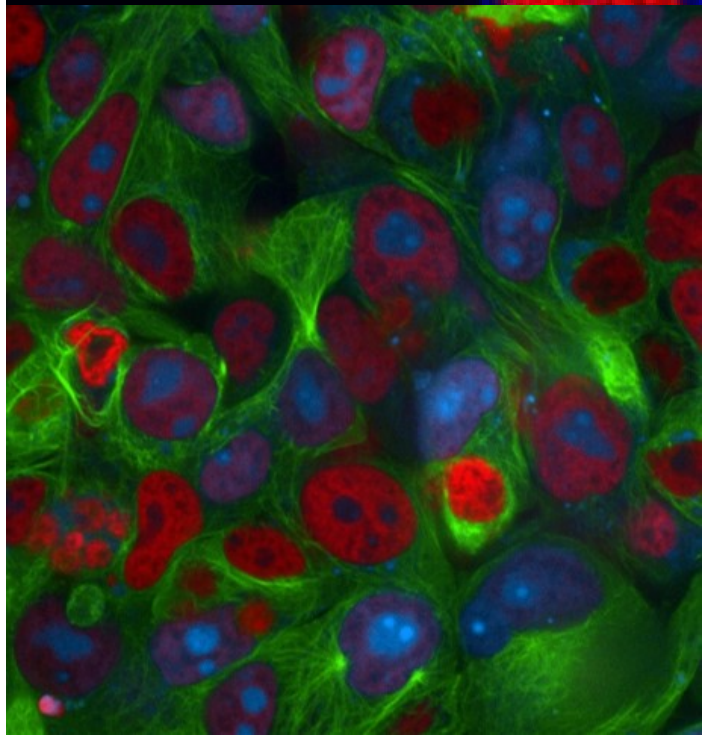
- SARS-CoV-2 Spike Protein
- SARS-CoV-2 Nucleocapsid Protein
- TMPRSS2
- ACE2
- BSG (CD147)
- SARS-CoV Nucleocapsid Protein
- SARS-CoV Spike Protein
- MERS-CoV Nucleocapsid Protein
- MERS-CoV Spike Protein

- Target delivery of first product lines: June
- To be available as polyclonal or monoclonal lines (polyclonal to be available earlier)
- Accepting orders now

Custom Cell Lines

Inserting Genes for Tagging or Overexpression





(1) Speed—Pure, Knock-in Custom Cell Lines up to 75% Faster; (2) Better Models—Up to 3 Knock-ins in the Same Cell Line

FAST-HDR

Create donor vector: 1 d

Isolate pure, modified cells: ≈ 10 d

Conventional methods

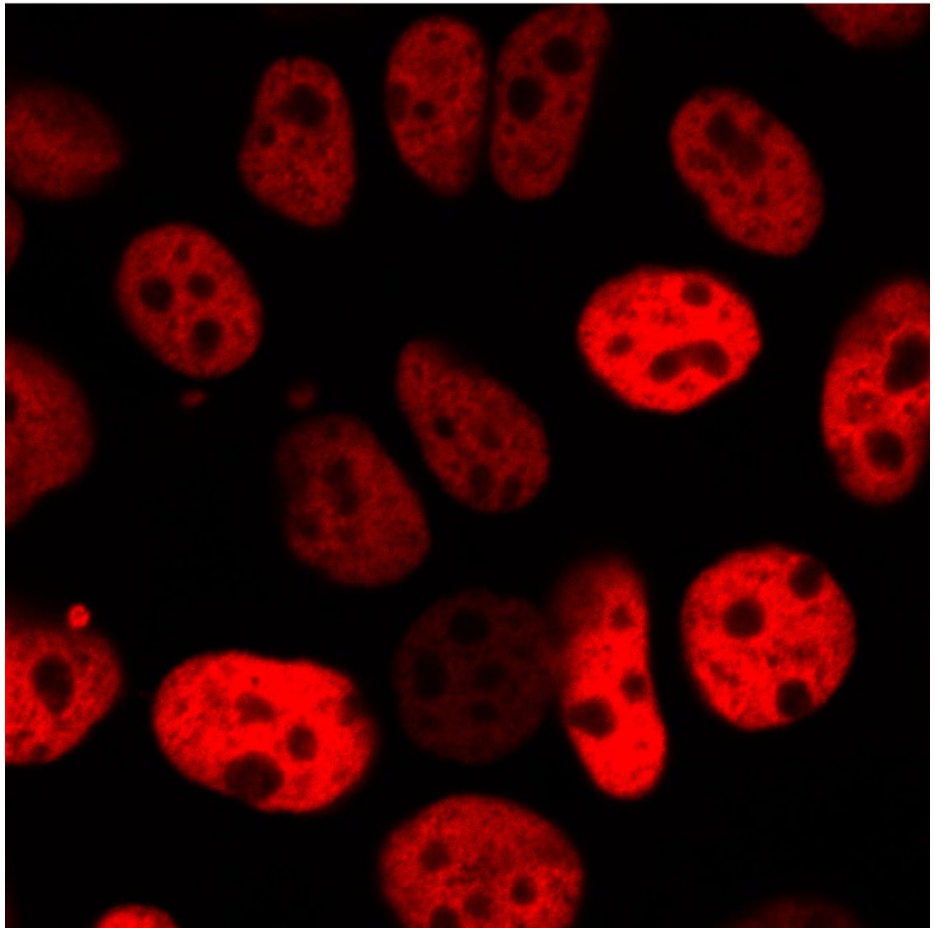
Create donor vector: 21 d

Isolate pure, modified cells: 4 – 8 weeks

Workflow efficiencies enable the creation and expansion of custom cell lines containing three reporter knock-ins in same time other companies insert one gene

Endogenous, Single Gene Labeling

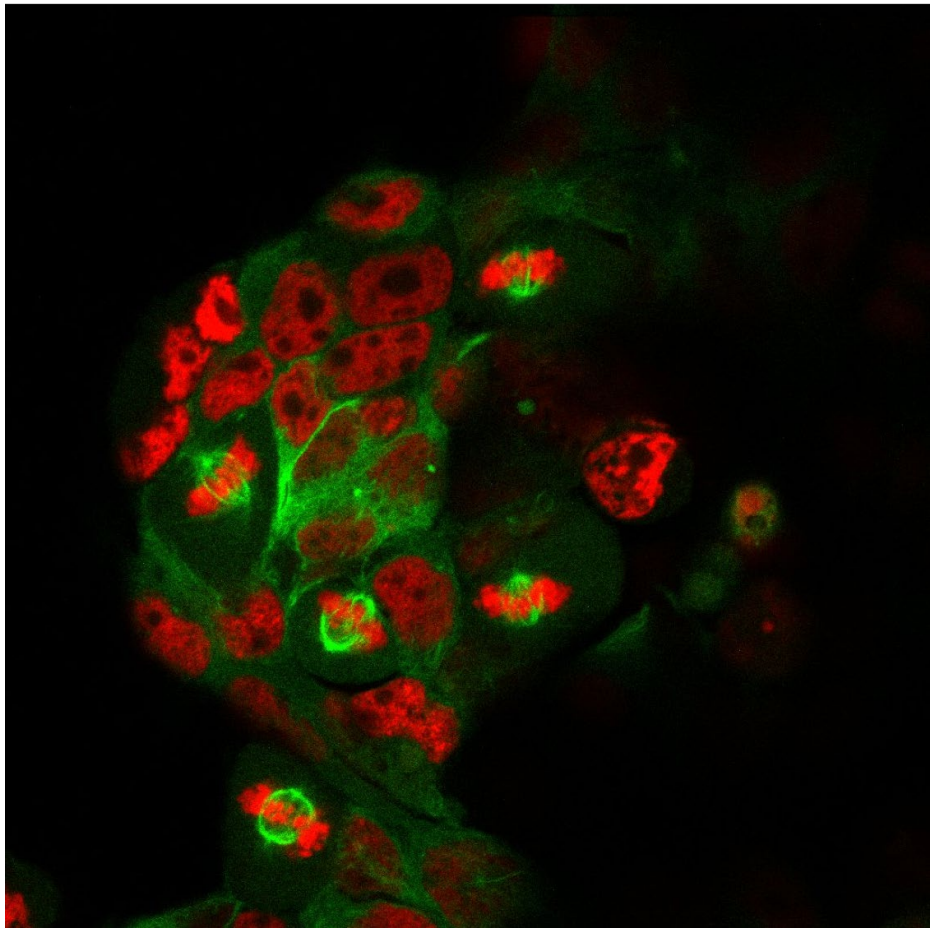
Tagging With mRuby3, Selected With Zeocin™, Day 14 Following Transfection



This image shows HEK293T cells in which endogenous Histone 3.3 has been tagged with mRuby3 and selected with Zeocin™. Image captured 14 days following transfection.

Double Gene Labeling

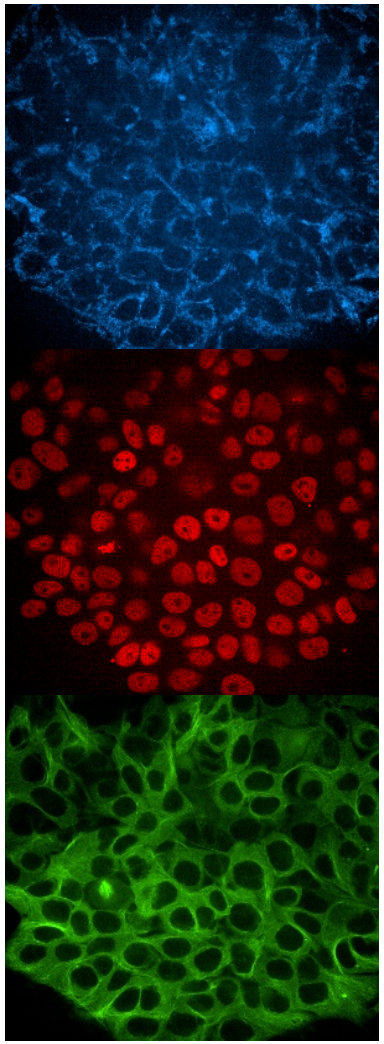
Tagging With mRuby3 + mClover, Selection With Zeocin™ + Puromycin

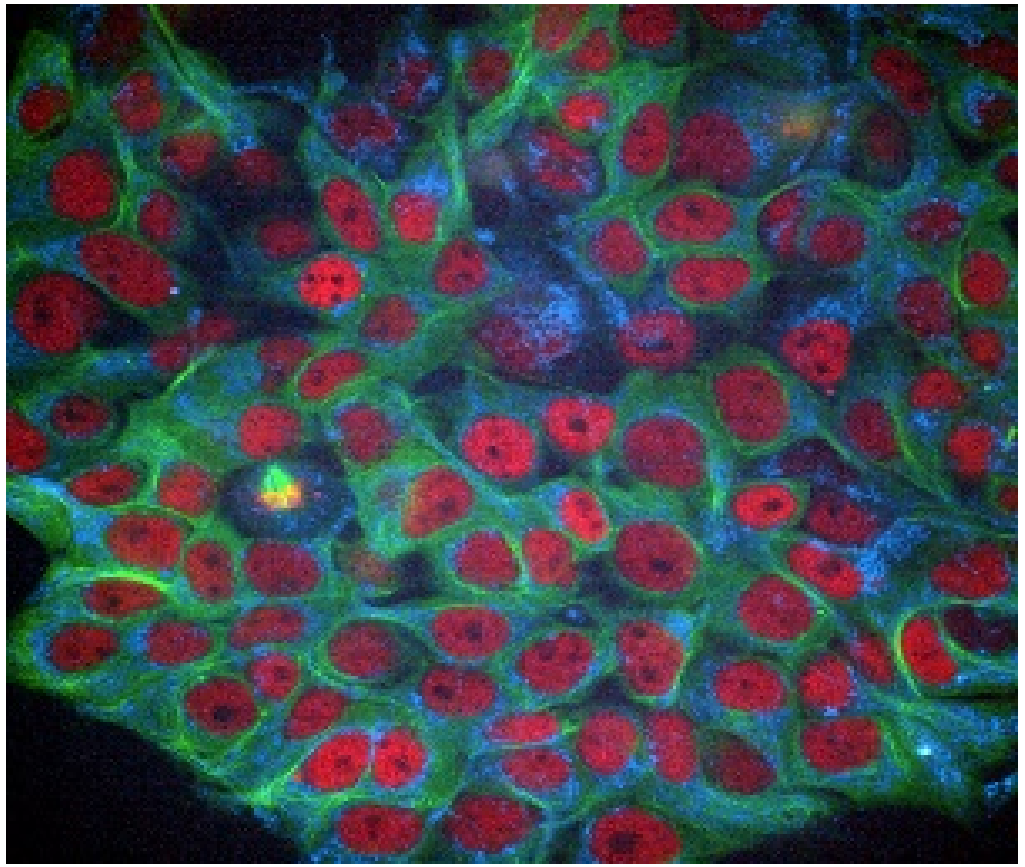


This image shows HEK293T cells in which endogenous Histone 3.3 has been tagged first with mRuby3 and subsequently β-tubulin has been tagged with mClover3. Cells were selected with a mix of Zeocin™ and puromycin. Image captured Day 14 following transfection.

Triple Gene Labeling

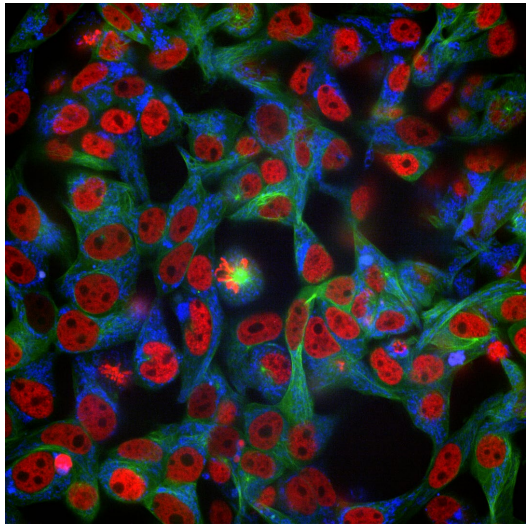
ATP5B Tagging With mTagBFP2, β-Tubulin With mClover3, Histone 3.3 With mRuby3





Endogenous ATP5B has been tagged with mTagBFP2, Histone 3.3 with mRuby3, and β-tubulin with mClover3. Cells selected with a mix of Zeocin™, puromycin, and blasticidin.

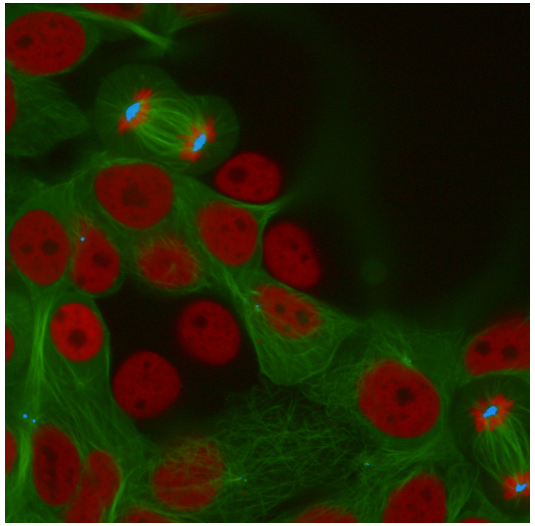
Benefit for the Researcher: Better Cell Models for Drug Discovery



Mitochondria

Red: H3.3-mRuby3

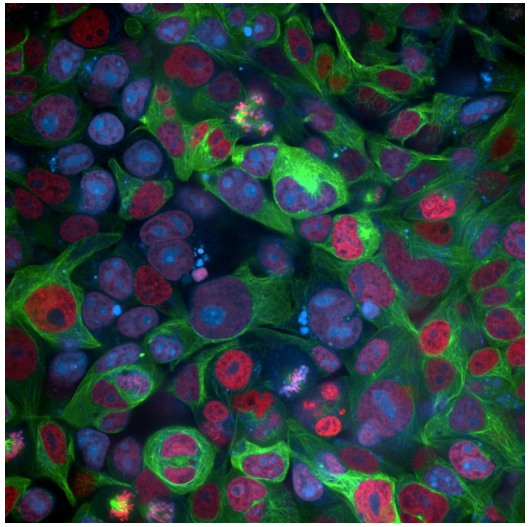
Green: β-Tubulin



Spindle poles

Red: H3.3-mRuby3

Green: β-Tubulin



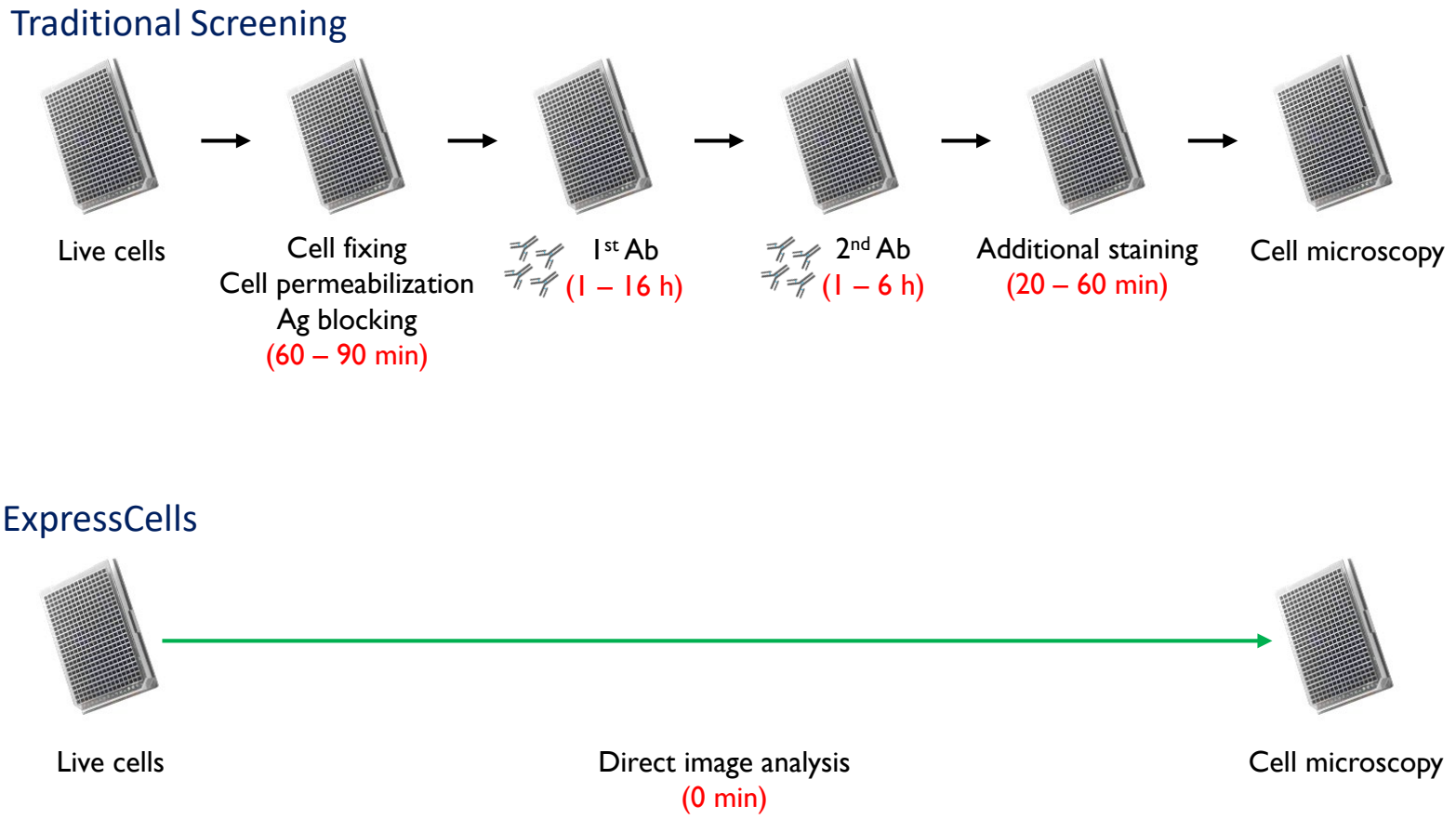
Nucleus

Red: H3.3-mRuby3

Green: β-Tubulin

Examples of live-cell microscopy without the use of staining or immuno-fluorescence. On the left, triple-labeling in which mitochondria (ATP5B) fluoresce in blue, Histone 3.3 in red, and β-tubulin in green. In the center, spindle poles during mitosis fluoresce in blue. On the right, dual staining of the nucleus. PARP1 is tagged in blue, Histone 3.3 in red.

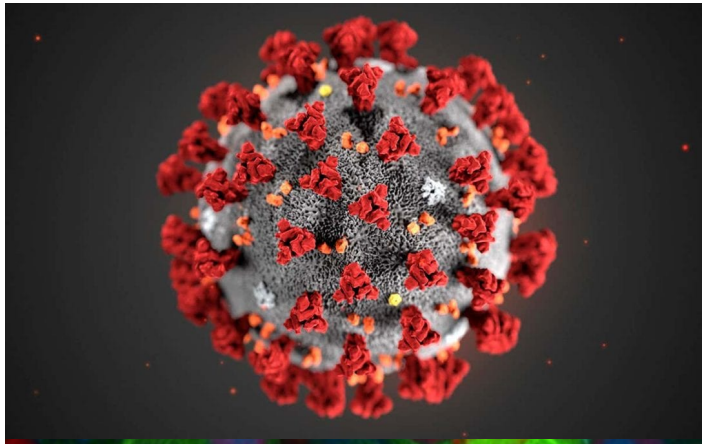
Benefit for the Researcher: Triple-Labeling With FAST-HDR Obviates the Need for Immunofluorescence and Staining

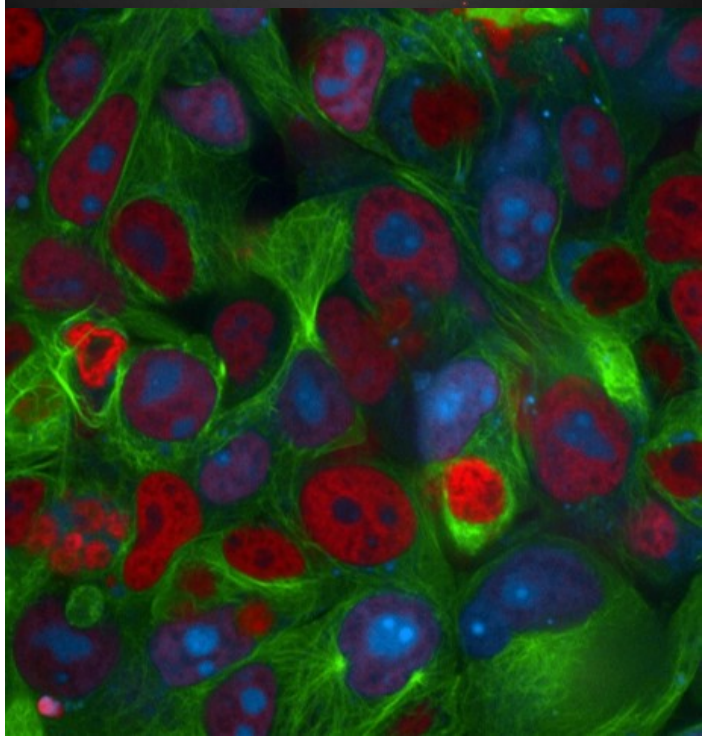


Track or study up to three different targets in live cells without labor-intensive staining or immunofluorescence.

Uses of ExpressCells Cell Lines

- Longitudinal screening
  - Compound library screening for better target- or phenotype-based hit identification
  - Cell-based toxicology assays: shrink time for ruling out drug candidates due to toxicity
  - Identify inhibitors of intra-cellular signaling pathways without complicated assays
- Discriminating among protein sequence variants
- Defining protein–membrane interactions
  - Reduces misleading results due to fixation and staining
- Rapid homozygous tagging of target genes





ExpressCells

Better knock-ins, better cell lines, better science.

Ready to Deliver Knock-In Cell Lines

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