# HAP1 knockout cell line application references.

The use of Revvity's HAP1 cell lines has expanded into a wide range of reaseach applications such as studies in virology, functional genomics, energy metabolism, and apoptosis, to name a few. Highlighted below is a list of peer-reviewed publicaitons that cited Revvity's HAP1 cell lines along with their corresponding functional assay methodologies.

## Virology

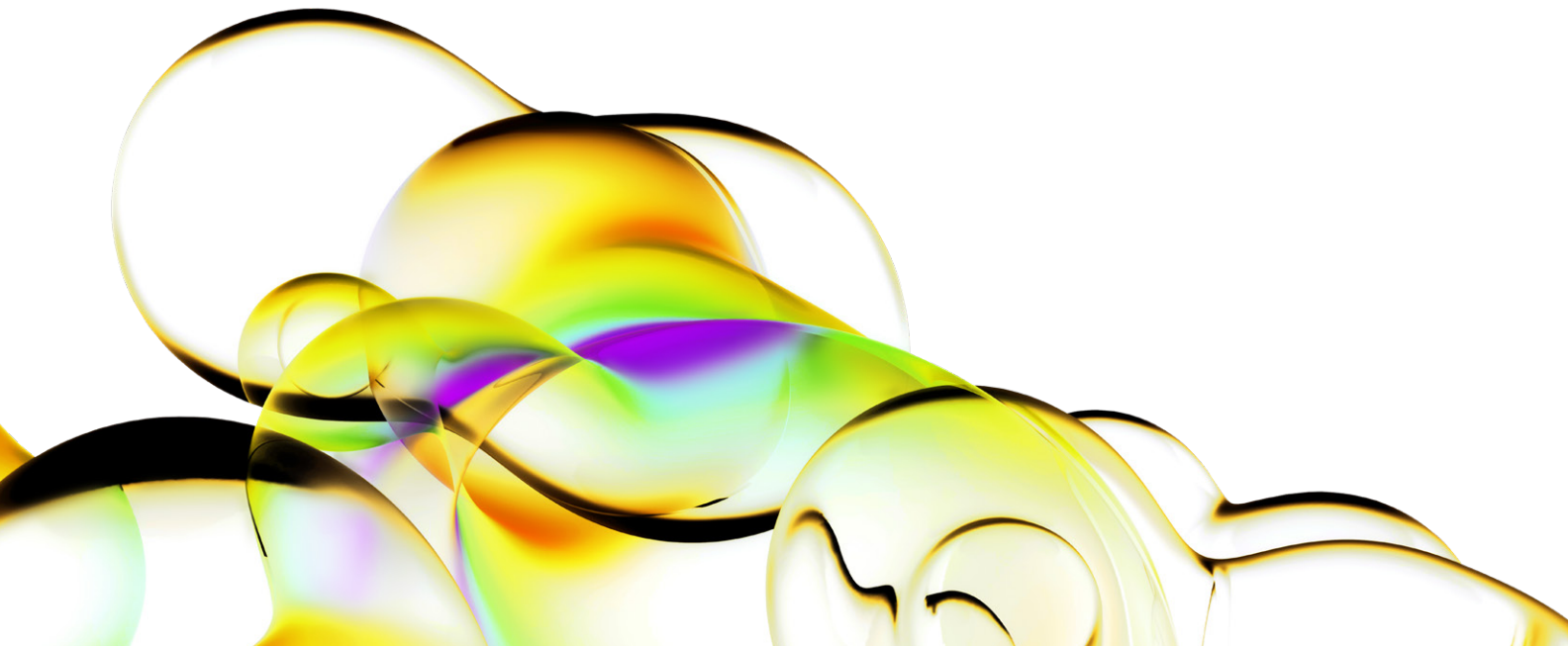

1. Flint, M. et al. A genome-wide CRISPR screen identifies N-acetylglucosamine-1-phosphate transferase as a potential antiviral target for Ebola virus. *Nat. Commun.* **10**, 285 (2019).

2. Lyoo, H. et al. ACBD3 Is an Essential Pan-enterovirus Host Factor That Mediates the Interaction between Viral 3A Protein and Cellular Protein PI4KB. *mBio* 10, e02742-18, /mbio/10/1/mBio.02742-18.atom (2019).

3. Baggen, J. et al. Bypassing pan-enterovirus host factor PLA2G16. *Nat. Commun.* 10, 3171 (2019).

4. McPhail, J. A. et al. Characterization of the Golgi c10orf76-PI4KB complex, and its necessity for Golgi PI4P levels and enterovirus replication. http://biorxiv.org/lookup/doi/10.1101/634592 (2019) doi:10.1101/634592.

5. Liu, Y. et al. Correction to: Tat expression led to increased histone 3 tri-methylation at lysine 27 and contributed to HIV latency in astrocytes through regulation of MeCP2 and Ezh2 expression. *J. Neurovirol.* 25, 901–901 (2019).

6. Pokharel, S. M. et al. Integrin activation by the lipid molecule 25-hydroxycholesterol induces a proinflammatory response. *Nat. Commun.* 10, 1482 (2019).

7. Shen, Q. et al. RanBP2/Nup358 enhances miRNA activity by sumoylating and stabilizing Argonaute 1. https://www.biorxiv.org/content/10.1101/555896v2.

8. LaFontaine, E. et al. Ribosomal protein RACK1 facilitates efficient translation of poliovirus and other viral IRESs. http://biorxiv.org/lookup/doi/10.1101/659185 (2019) doi:10.1101/659185.

9. Moskovskich, A. et al. The transporters SLC35A1 and SLC30A1 play opposite roles in cell survival upon VSV virus infection. http://biorxiv.org/lookup/doi/10.1101/573253 (2019) doi:10.1101/573253.

10. Chiramel, A. I. et al. TRIM5α Restricts Flavivirus Replication by Targeting the Viral Protease for Proteasomal Degradation. *Cell Rep.* 27, 3269-3283.e6 (2019).

**Methods:** CellTiter-Glo viability assays (Promega, Madison, USA)[1]; reporter virus assays[1]; virus quantification (endpoint dilution)[2]; luciferase reporter assays[8]; plaque diameter/size assays[8]; NFκB measurement[6]. Golgi staining[4]; Viral time-course assays[9]; Zn-level quantification[9]; Cell Death/Apoptosis assays[9]; IL6 quantification[7].


## Genome integrity/Functional genomics


1. Bacolla, A., Ye, Z., Ahmed, Z. & Tainer, J. A. Cancer mutational burden is shaped by G4 DNA, replication stress and mitochondrial dysfunction. *Prog. Biophys. Mol. Biol.* 147, 47-61 (2019).

2. Cui, J., Gizzi, A. & Stivers, J. T. Deoxyuridine in DNA has an inhibitory and promutagenic effect on RNA transcription by diverse RNA polymerases. *Nucleic Acids Res.* (2019) doi:10.1093/nar/gkz183.

3. Serebrenik, Y. V., Sansbury, S. E., Kumar, S. S., Henao-Mejia, J. & Shalem, O. Efficient and flexible tagging of endogenous genes by homology-independent intron targeting. *Genome Res.* 29,1322–1328 (2019).

4. Mair, B. et al. Essential Gene Profiles for Human Pluripotent Stem Cells Identify Uncharacterized Genes and Substrate Dependencies. *Cell Rep.* 27, 599-615.e12 (2019).

5. Xing, M. & Oksenych, V. Genetic interaction between DNA repair factors PAXX, XLF, XRCC4 and DNA–PKcs in human cells. *FEBS Open Bio.* 9, 1315–1326 (2019).

6. Castañe da-Zegarra, S., Xing, M., Gago-Fuentes, R., Sæterstad, S. & Oksenych, V. Synthetic lethality between DNA repair factors Xlf and Paxx is rescued by inactivation of Trp53. *DNA Repair* 73, 164–169 (2019).

7. Garvin, A. J. et al. The deSUMOylase SENP2 coordinates homologous recombination and nonhomologous end joining by independent mechanisms. *Genes Dev.* 33, 333–347 (2019).

8. Sarno, A. et al. Uracil–DNA glycosylase UNG1 isoform variant supports class switch recombination and repairs nuclear genomic uracil. *Nucleic Acids Res.* 47, 4569–4585 (2019).

**Methods:** G4 DNA staining[1]; Intron tagging[3]; survival assays of NHEJ factors deficient HAP1 cells[5]; cell cycle synchronization/staining[7].


## Mitochondria/Energy metabolism


1. Sánchez-Caballero, L. et al. A dual function of TMEM70 in OXPHOS: assembly of complexes I and V. http://biorxiv.org/lookup/doi/10.1101/697185 (2019) doi:10.1101/697185.

2. Kondadi, A. K. et al. Cristae undergo continuous cycles of fusion and fission in a MICOS-dependent manner. http://biorxiv.org/lookup/doi/10.1101/654541 (2019) doi:10.1101/654541.

3. Yang, Y., Mohammed, F. S., Zhang, N. & Sauve, A. A. Dihydronicotinamide riboside is a potent NAD + concentration enhancer *in vitro* and *in vivo*. *J. Biol. Chem.* 294, 9295–9307 (2019).

4. Małecki, J. M. et al. Human FAM173A is a mitochondrial lysine-specific methyltransferase that targets adenine nucleotide translocase and affects mitochondrial respiration. *J. Biol. Chem.* 294, 11654–11664 (2019).

5. Małecki, J. M. et al. Lysine methylation by the mitochondrial methyltransferase FAM173B optimizes the function of mitochondrial ATP synthase. *J. Biol. Chem.* 294, 1128–1141 (2019).

6. Gioran, A. et al. Multi–omics identify xanthine as a pro-survival metabolite for nematodes with mitochondrial dysfunction. *EMBO J.* 38, (2019).

7. Guiducci, G. et al. The moonlighting RNA-binding activity of cytosolic serine hydroxymethyltransferase contributes to control compartmentalization of serine metabolism. *Nucleic Acids Res.* 47, 4240–4254 (2019).

**Methods:** electron microscopy[2], super-resolution nanoscopy[6]; respiration measurements by Seahorse [2,4,5,6]; NADH measurement[3]; detection of respiratory chain complexes (MS/MS analysis)[1,4]; Pulse labelling of mitochondrial translation products[1]; cell lysate activity tests of KO/WT cells[3].

## Autophagy/Apoptosis

1. Cao, J. Y. et al. A Genome-wide Haploid Genetic Screen Identifies Regulators of Glutathione Abundance and Ferroptosis Sensitivity. *Cell Rep.* 26, 1544-1556.e8 (2019).

2. Lenk, G. M. et al. CRISPR knockout screen implicates three genes in lysosome function. *Sci. Rep.* 9, 9609 (2019).

3. Keskitalo, S. et al. Dominant TOM1 mutation associated with combined immunodeficiency and autoimmune disease. *Npj Genomic Med.* 4, 14 (2019).

4. Atakpa, P., van Marrewijk, L. M., Apta-Smith, M., Chakraborty, S. & Taylor, C. W. GPN does not release lysosomal Ca 2+ but evokes Ca 2+ release from the ER by increasing the cytosolic pH independently of cathepsin C. J. *Cell Sci.* 132, jcs223883 (2019).

5. Agrotis, A., Pengo, N., Burden, J. J. & Ketteler, R. Redundancy of human ATG4 protease isoforms in autophagy and LC3/GABARAP processing revealed in cells. *Autophagy* 15, 976–997 (2019).

6. Simons, I. M. et al. The highly GABARAP specific rat monoclonal antibody 8H5 visualizes GABARAP in immunofluorescence imaging at endogenous levels. *Sci. Rep.* 9, 526 (2019).

**Methods**: human haploid cell genetic screening[1]; glutathione (GSH) quantification via monochlorobimane (MCB) GSH probes/FACS Analysis and Ellman's reagent[1]; immuno-staining via autophagy marker (e.g.LC3 puncta)[5]; transmission electron microscopy (TEM)[5].

## Drug validation

1. Hopkins, T. A. et al. PARP1 Trapping by PARP Inhibitors Drives Cytotoxicity in Both Cancer Cells and Healthy Bone Marrow. *Mol. Cancer Res.* 17, 409–419 (2019).

2. Depetter, Y. et al. Selective pharmacological inhibitors of HDAC6 reveal biochemical activity but functional tolerance in cancer models: Activity of HDAC6 inhibitors in cancer models. *Int. J. Cancer.* 145, 735–747 (2019).

**Methods**: drug cytotoxicity assays[1]; drug target validation[2].

## Extracellular matrix (ECM)

1. Blum, A. et al. Transcriptomics of a KDELR1 knockout cell line reveals modulated cell adhesion properties. *Sci. Rep.* 9, 10611 (2019).

**Methods:** whole transcriptome analysis, *in vitro* adhesion assays (e.G. scratch assays).

## Ubiquitylation

1. Campagne, A. et al. BAP1 complex promotes transcription by opposing PRC1-mediated H2A ubiquitylation. *Nat. Commun.* 10, 348 (2019).

**Methods:** chromatography analysis of nuclear extracts.

## Epigenetics

1. Choi, S. et al. H2A.Z-dependent and -independent recruitment of metabolic enzymes to chromatin required for histone modifications. http://biorxiv.org/lookup/doi/10.1101/553297 (2019) doi:10.1101/553297.

**Methods:** cellular extraction in cytosol, mitochondrial and membrane, nuclear, and chromatin-bound protein fractions.

## Translational apparatus


1. Vindry, C., Guillin, O., Mangeot, P. E., Ohlmann, T. & Chavatte, L. A Versatile Strategy to Reduce UGA-Selenocysteine Recoding Efficiency of the Ribosome Using CRISPR-Cas9-Viral-Like-Particles Targeting Selenocysteine-tRNA[Ser]Sec Gene. *Cells* 8, 574 (2019).

2. Henriques, S. F., Gicquel, E., Marsolier, J. & Richard, I. Functional and cellular localization diversity associated with Fukutin–related protein patient genetic variants. Hum. *Mutat*. 40, 1874–1885 (2019).


**Methods:** Northern blot analysis[1], tRNA transduction[1]; TIDE analysis[1].

## Cell biology


1. Nixon, A. M. L. et al. A rapid *in vitro* methodology for simultaneous target discovery and antibody generation against functional cell subpopulations. *Sci. Rep*. 9, 842 (2019).

2. Sergeeva, O. A. & van der Goot, F. G. Anthrax toxin requires ZDHHC5- mediated palmitoylation of its surface-processing host enzymes. *Proc. Natl. Acad. Sci.* 116, 1279–1288 (2019).

3. Taneja, N. & Burnette, D. T. Myosin IIA drives membrane bleb retraction. *Mol. Biol. Cell* 30, 1051–1059 (2019).

4. Taneja, N. et al. Precise tuning of cortical contractility regulates cell shape during cytokinesis. http://biorxiv.org/lookup/doi/10.1101/635615 (2019) doi:10.1101/635615.

5. Tuladhar, R. et al. Stereoselective fatty acylation is essential for the release of lipidated WNT proteins from the acyltransferase Porcupine (PORCN). *J. Biol. Chem*. 294, 6273–6282 (2019).

6. Karsai, G. et al. DEGS1-associated aberrant sphingolipid metabolism impairs nervous system function in humans. *J. Clin. Invest*. 129, 1229–1239 (2019).


**Methods:** differential Interference Contrast (DIC) microscopy and Fluorescence Recovery After Photobleaching (FRAP)[4]; WNT secretion assay (cdDNA transfection, ConA-Sepharose affinity chromatography and Western Blot Analysis)[5]; metabolite labeling and detection[6].

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