

**Datasheet for 451Lu-01-0001****451Lu Viable Cells****Overview**

|                      |                                    |
|----------------------|------------------------------------|
| <b>Description:</b>  | 451Lu Viable Cells - 451Lu-01-0001 |
| <b>Item No.:</b>     | 451Lu-01-0001                      |
| <b>Size:</b>         | 1 million cells                    |
| <b>Applications:</b> | Cellular Assay, Other, WB          |
| <b>Origin:</b>       | Human                              |

**Product Details**

|                           |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                |
|---------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Background:</b>        | 451Lu is a selected xenograft aggressive metastatic sub-line of WM164. WM164 is a metastatic human melanoma cell line established from a metastatic site (right upper arm) in a 22-year-old male with stage IV superficial spreading melanoma. 451Lu cell line features the specific V600E (Val600Glu) mutation at codon 600 in the BRAF gene. This mutation causes constitutively active kinase activity and activation of MEK and ERK signaling pathway. This cell line is wild type for PTEN, N-RAS, c-KIT, and CDK4 genes. |
| <b>Synonyms:</b>          | Melanoma, patient derived tumor, tumor models, skin cancer, xenograft                                                                                                                                                                                                                                                                                                                                                                                                                                                          |
| <b>Species of Origin:</b> | Human                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                          |

**Target Details**

|                            |                                                                                                                                                                                                                                                                                                                                           |
|----------------------------|-------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Purity/Specificity:</b> | Cells are sterile, validated by short tandem repeat profiling, and are tested as negative for mycoplasma. It is recommended that cell lines are tested for mycoplasma contamination and short tandem repeat (STR) profiling every 10 passages or each time a frozen seed stock is made. See cell culture protocol for additional details. |
| <b>Relevant Links:</b>     | <ul style="list-style-type: none"><li>• <a href="#">Cell Line EULA</a></li><li>• <a href="#">Melanoma Cell Culture Protocol</a></li></ul>                                                                                                                                                                                                 |

**Application Details**

|                                |                                                 |
|--------------------------------|-------------------------------------------------|
| <b>Suggested Applications:</b> | Cellular Assay, Other, WB (Based on references) |
|--------------------------------|-------------------------------------------------|

**Application Note:** The key applications of these cell lines include genetic studies, xenograft production, drug testing, and drug target discovery. These cell line models can be used in various biological assays, and for identifying critical target genes, and cell signaling pathways.

**Assay Dilutions:** All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.

## Cell Line Data

|                        |                                                                                                                                                              |
|------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Cell Line:</b>      | Human Melanoma                                                                                                                                               |
| <b>Product Type:</b>   | Viable Cells                                                                                                                                                 |
| <b>Cell Viability:</b> | Yes                                                                                                                                                          |
| <b>Stage:</b>          | Metastasis                                                                                                                                                   |
| <b>BRAF:</b>           | V600E                                                                                                                                                        |
| <b>CDK4:</b>           | WT                                                                                                                                                           |
| <b>C-Kit:</b>          | WT                                                                                                                                                           |
| <b>N-RAS:</b>          | WT                                                                                                                                                           |
| <b>PTEN:</b>           | WT                                                                                                                                                           |
| <b>Paired:</b>         | Yes                                                                                                                                                          |
| <b>Medium:</b>         | Tumor Specialized Media with 2% HI-FBS                                                                                                                       |
| <b>Sub-culture:</b>    | Cells should be maintained between 30 – 95% confluence in tumor specialized medium with 2% FBS; split cultures 1:10 every 5-6 days using 0.25% trypsin/EDTA. |
| <b>Incubation:</b>     | 36°C with 5% CO <sub>2</sub>                                                                                                                                 |

## Formulation

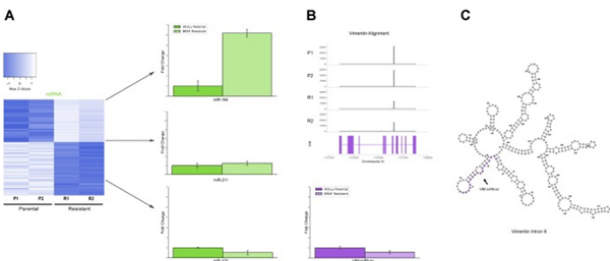
|                        |                                                   |
|------------------------|---------------------------------------------------|
| <b>Physical State:</b> | Frozen Cell Suspension                            |
| <b>Concentration:</b>  | 1x10 <sup>6</sup> cells/mL Count By Hemocytometer |
| <b>Buffer:</b>         | None                                              |
| <b>Preservative:</b>   | None                                              |
| <b>Stabilizer:</b>     | None                                              |

## Shipping & Handling

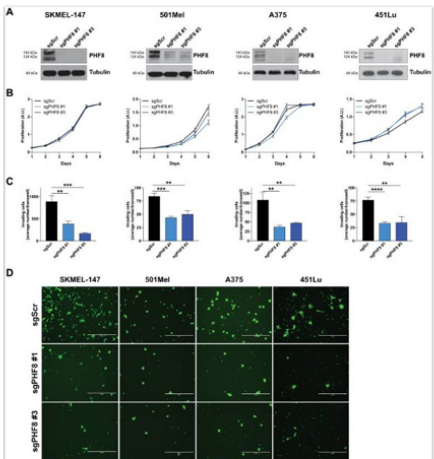
**Shipping Condition:** Dry Ice

|                    |                                                                                                                                  |
|--------------------|----------------------------------------------------------------------------------------------------------------------------------|
| Storage Condition: | Cells are frozen with 90% FBS/10% DMSO solution at about 1x10 <sup>6</sup> cells/ml. Store vial in liquid nitrogen upon arrival. |
| Expiration:        | Expiration date is two (2) years from date of receipt.                                                                           |

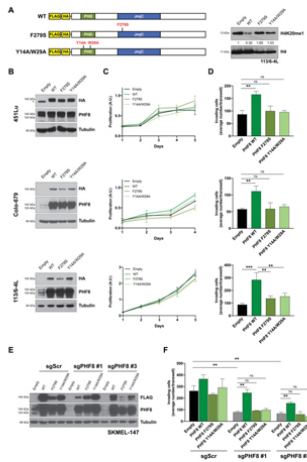
Images



**Figure**  
Detailed analysis miRNAs of interest. (A) qPCR representing fold change of miRNAs miR-184, miR-211, and miR-100 in 451Lu BRAF Resistant samples relative to 451Lu parental expression levels across replicates. (B) qPCR representing log fold change of the VIM miRNA in 451Lu BRAF Resistant vs. Parental samples and BAM gene alignment tracks across samples. (C) RNAfold predicted secondary structure of VIM intron 6 with the candidate miRNA highlighted in purple. Fig 4. PMID: 30349559

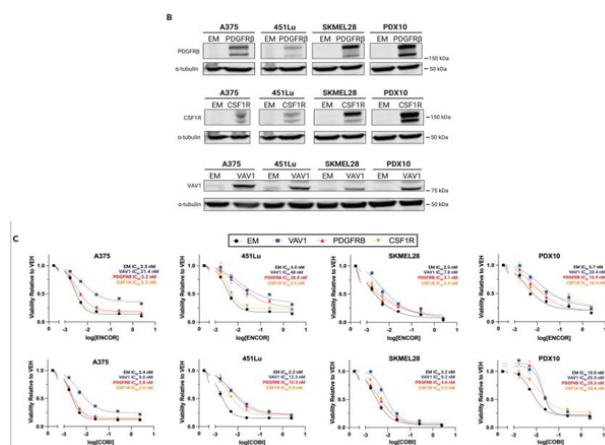


**Western Blot**  
(A) Efficient PHF8 knockout using two different sgRNAs in four different melanoma cell lines was determined by Western blot. The membrane was reblotted with tubulin antibody for loading control. (B) Proliferation curves of cells seeded 3 days after infection with sgRNA-carrying lentiviral particles targeting PHF8. (C) Invasion assay of cells seeded 3 days after infection with sgRNA-containing lentiviral particles, using Fluoroblok transwell chambers coated with Matrigel. PHF8 knockout significantly inhibits melanoma invasion in SKMEL-147 (sgPHF8 #1 versus sgScr, P = 0.003; sgPHF8 #3 versus sgScr, P = 0.0007), 501Mel (sgPHF8 #1 versus sgScr, P = 0.0003; sgPHF8 #3 versus sgScr, P = 0.002), A375 (sgPHF8 #1 versus sgScr, P = 0.003; sgPHF8 #3 versus sgScr, P = 0.006), and 451Lu cells (sgPHF8 #1 versus sgScr, P = 0.0001; sgPHF8 #3 versus sgScr, P = 0.003). (D) Representative pictures of the Fluoroblok transwell inserts stained with Calcein AM to quantify invading cells in (C). Scale bars, 400  $\mu$ m. Fig 2. PMID: 35179962



### Western Blot

(A) Left: Lentiviral constructs encoding for FLAG-HA tagged PHF8 WT, F279S, and Y14A/W29A. Right: Immunoblotting of H4K20me1 in 113/6-4L overexpressing PHF8 WT or PHF8 mutants. H4 Western blot serves as loading control. Band intensity relative to Empty condition is shown. (B) Efficient overexpression of PHF8 WT and mutant constructs in three cell lines was determined by Western blot using HA and PHF8 antibodies. (C) Proliferation and (D) Invasion assays were performed in 451Lu (top: PHF8 WT versus Empty,  $P = 0.002$ ; PHF8 F279S versus Empty,  $P = 0.42$ ; PHF8 Y14A/W29A versus Empty,  $P = 0.43$ ), Colo-679 (middle: PHF8 WT versus Empty,  $P = 0.004$ ; PHF8 F279S versus Empty,  $P = 0.91$ ; PHF8 Y14A/W29A versus Empty,  $P = 0.13$ ), and 113/6-4L cells (bottom: PHF8 WT versus Empty,  $P = 0.0004$ ; PHF8 F279S versus PHF8 WT,  $P = 0.002$ ; PHF8 Y14A/W29A versus PHF8 WT,  $P = 0.005$ ). (E) Efficient overexpression of PHF8 WT and mutant constructs in SKMEL-147 cells previously transduced with sgScr or sgPHF8 #3 was determined by Western blot. (F) Invasion assay shows that ectopic PHF8 WT expression, but not the mutant constructs, restores the invasive potential of PHF8 knockout SKMEL-147 cells (sgPHF8 #1 + Empty versus sgScr + Empty,  $P = 0.005$ ; sgPHF8 #1 + PHF8 WT versus sgPHF8 #1 + Empty,  $P = 0.002$ ; sgPHF8 #1 + PHF8 F279S versus sgPHF8 #1 + Empty,  $P = 0.14$ ; sgPHF8 #1 + PHF8 Y14A/W29A versus sgPHF8 #1 + Empty,  $P = 0.089$ ; sgPHF8 #3 + Empty versus sgScr + Empty,  $P = 0.004$ ; sgPHF8 #3 + PHF8 WT versus sgPHF8 #3 + Empty,  $P = 0.004$ ; sgPHF8 #3 + PHF8 F279S versus sgPHF8 #3 + Empty,  $P = 0.69$ ; sgPHF8 #3 + PHF8 Y14A/W29A versus sgPHF8 #3 + Empty,  $P = 0.098$ ). Error bars indicate average  $\pm$  SD. Representative data of three independent experiments conducted are shown. Western blot membranes were reblotted with tubulin antibody for loading control. Fig 5. PMID: 35179962

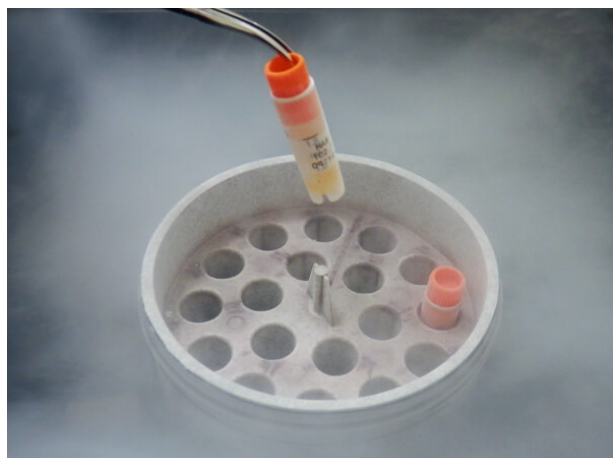


## Western Blot

(B) Representative western blot with indicated antibodies in a panel of melanoma cell lines engineered to overexpress the screen candidates, empty vector (EM), VAV1, PDGFRB, or CSF1R.

(C) Dose-response of modified cell lines against ENCOR and COBI. Top row shows ENCOR data and bottom row shows COBI data. Each data point represents three replicate measurements. Viability is shown after three days of drug treatment for A375 and 451Lu and four days for SKMEL28 and PDX10. The error bars denote the 95% confidence interval.

Fig 3. PMID: 37860756



## Flask

Human melanoma tumor cells with known gene mutations, disease stage, STR, and RPPA profiling

## References

- Eller S et al. Exploiting metabolic adaptations to overcome dabrafenib treatment resistance in melanoma cells. *Mol Oncol.* (2026)
- Zhu EY et al. Understanding cancer drug resistance with Sleeping Beauty functional genomic screens: Application to MAPK inhibition in cutaneous melanoma. *iScience* (2023)
- Bindi B et al. Bioinspired Collagen/Hyaluronic Acid/Fibrin-Based Hydrogels for Soft Tissue Engineering: Design, Synthesis, and In Vitro Characterization. *J Funct Biomater.* (2023)
- Moubarak RS et al. The histone demethylase PHF8 regulates TGF $\beta$  signaling and promotes melanoma metastasis. *Meta-Analysis.* (2022)
- Hanniford D et al. Epigenetic silencing of CDR1as drives IGF2BP3-mediated melanoma invasion and metastasis. *Cancer Cell.* (2021)
- O'Neill K et al. TEsma1 Identifies Small RNAs Associated With Targeted Inhibitor Resistance in Melanoma. *Front Genet.* (2018)

## Disclaimer

No test method can provide total assurance that the hepatitis B virus, hepatitis C virus, human immunodeficiency virus, or any other infectious agents are absent. Thus, all blood products, including purified proteins derived from human blood sources, should be handled at Biosafety Level 2 as recommended by the CDC\NIH manual entitled Biosafety in Microbiological and Biomedical Laboratories for potentially infectious human serum, blood specimens or proteins derived from same. Source material for the human blood product supplied to your facility has been tested for the detection of HIV antibody, Hepatitis B surface antigen, antibody to Hepatitis C, HIV 1 antigen(s), antibody to HTLV - I/II, and syphilis by FDA guidelines. All units were found to be non-reactive/negative for these tests. All human blood source material is collected in FDA licensed centers and is tested with FDA approved test kits.

Cell Line Limited Use License Required. THIS PRODUCT IS SUBJECT TO AN END-USER LICENSE AGREEMENT (EULA). BY ACCEPTING THIS PRODUCT, RECIPIENT AGREES TO BE BOUND BY THE TERMS OF USE SET FORTH BELOW and SET FORTH IN THE EULA. THIS PRODUCT IS FOR IN VITRO RESEARCH USE ONLY. THERAPEUTIC, DIAGNOSTIC, OR VETERINARY USE IS PROHIBITED. This product may not be resold or transferred by the recipient and may be used only by the recipient, in the recipient's facility and only for research use and other uses specifically permitted by the EULA. No other commercial use is allowed. "Commercial Use" means any and all uses of this product by recipient or others for monetary or other consideration, including providing services, supplying information or data to unaffiliated third parties, and resale or transfer of this product for any use. Recipient has no right to modify, derivatize, genetically engineer or otherwise create variations of this product or associated cells or cell lines. ROCKLAND AND WISTAR MAKE NO REPRESENTATIONS AND EXTEND NO WARRANTIES OF ANY KIND, EITHER EXPRESSED OR IMPLIED, INCLUDING AS TO MERCHANTABILITY OR FITNESS FOR A PARTICULAR PURPOSE, OR THAT THE USE OF THE PRODUCTS WILL NOT INFRINGE ANY PATENT, COPYRIGHT, TRADEMARK, OR OTHER PROPRIETARY RIGHTS. The terms set forth herein and in the EULA shall be governed by the laws of the Commonwealth of Pennsylvania, USA. To obtain a COMMERCIAL USE license for this product, please contact Rockland Immunochemicals, Inc. Please contact a technical service representative for more information. All properties listed are typical characteristics and are not specifications. All suggestions and data are offered in good faith but without guarantee as conditions and methods of use of our products are beyond our control. All claims must be made within 30 days following the date of delivery. The prospective user must determine the suitability of our materials before adopting them on a commercial scale. Suggested uses of our products are not recommendations to use our products in violation of any patent or as a license under any patent of Rockland Immunochemicals, Inc. If you require a commercial license to use this material and do not have one, then return this material, unopened to: Rockland Inc., P.O. BOX 5199, Limerick, Pennsylvania, USA.