**Supplementary Data**

# Comparative analysis of SEC61A1 mutant R236C in two patient-derived cellular platforms

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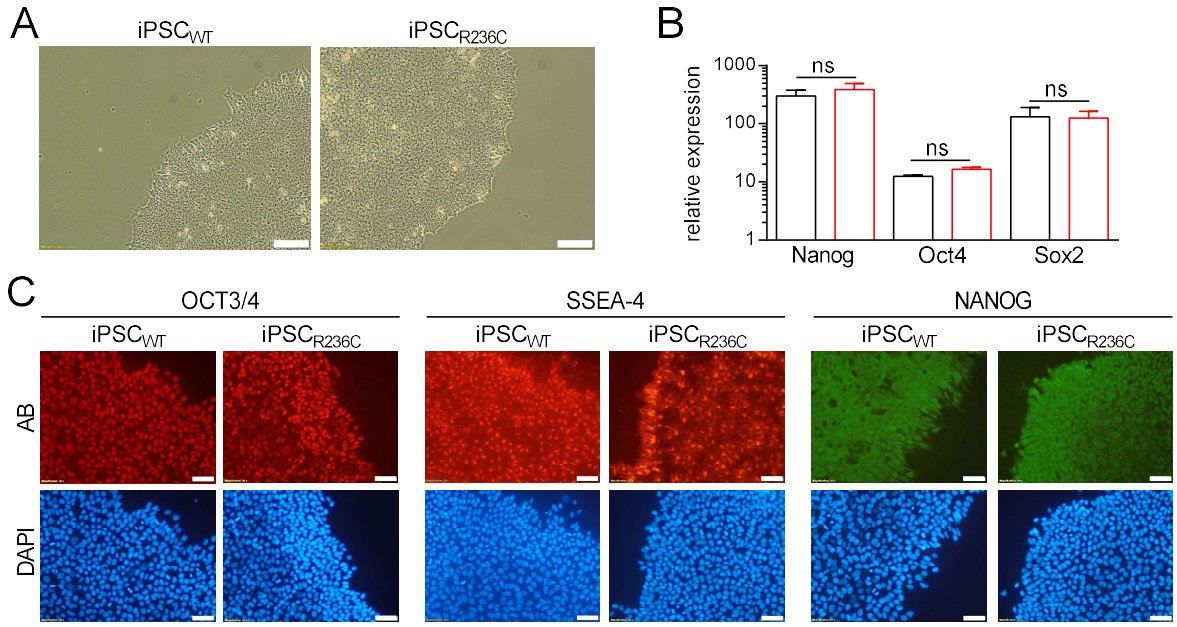
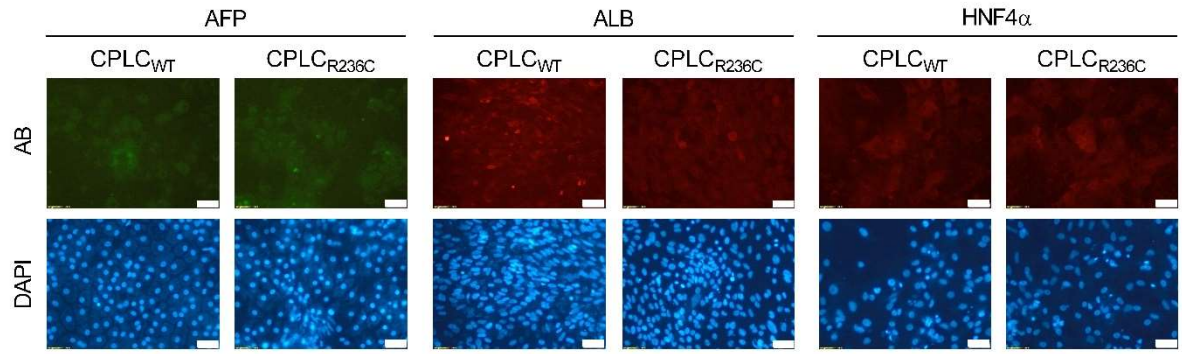


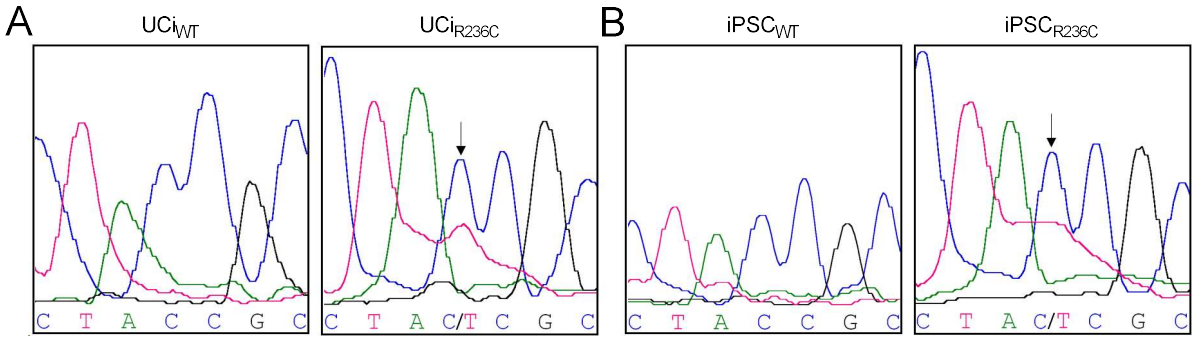

**Supplementary Figure S1.**

**iPSC derived from patient encoding R236C showed typical morphology and stem cell marker expression** (A) Typical bright field image. One of four experiments is shown. Magnification 10x. Scale bars, 100 µm. (B) Expression of stem cell marker genes derived from wildtype (black) and R236C cells (red). Mean±SE of relative expression to GAPDH is shown (n=4). (C) Immunfluorescence staining of cells against stem cell markers. DAPI was used for nuclear counterstaining. One of four experiments is shown. Magnification 20x. Scale bars, 50 µm. Mann-Whitney U test was used to test for statistical significance. ns, not significant p>0.05.



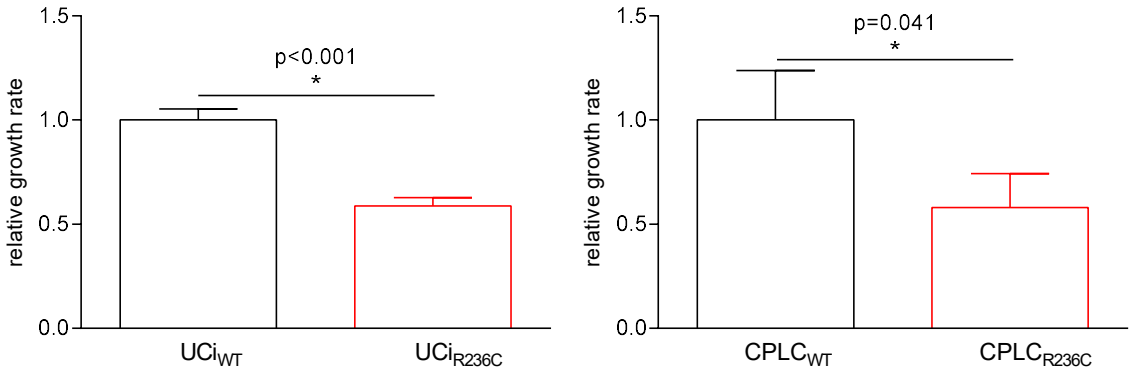

**Supplementary Figure S2.**

**Immunofluorescence of CPLCs showed expression of hepatocyte markers AFP, ALB and HNF4α.** DAPI was used for nuclear counterstaining. One of four experiments is shown. Magnification 20x. Scale bars, 50 μm.



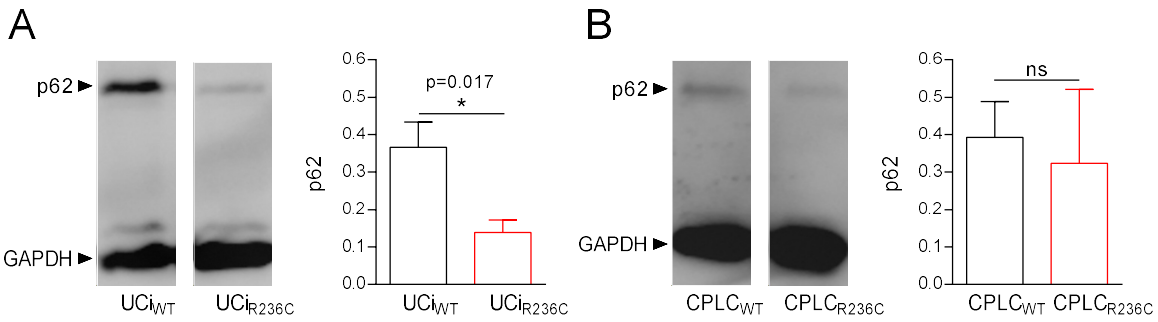

**Supplementary Figure S3.**

**Genotype c.706C>T (R236C) was maintained following retroviral immortalization (A) and reprogramming to iPSCs (B).** Chromatograms were established following sequence analyses of the *SEC61A1* gene. Heterozygotes at position 236 are indicated by arrow.



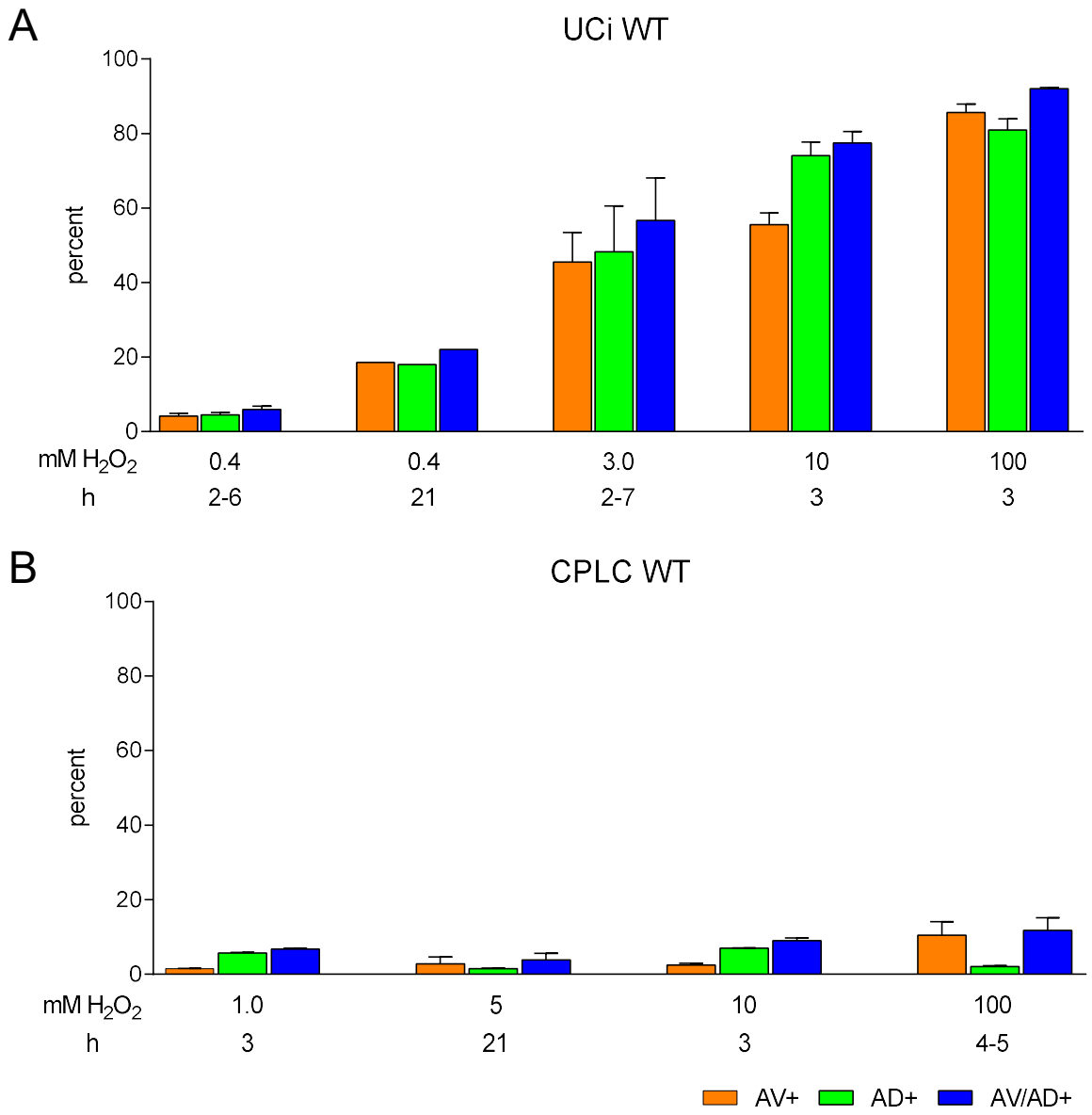

**Supplementary Figure S4.**

**Cells expressing R236C show reduced growth rate.** Cells were seeded at identical number at day 1. UCi growth rate was determined at day 3 after seeding (n=21). CPLC growth rate was determined at the end of differentiation at day 14 (n=6). The growth rate relative to WT is shown. Mann-Whitney U test was used to test for statistical significance. ns, not significant p>0.05.



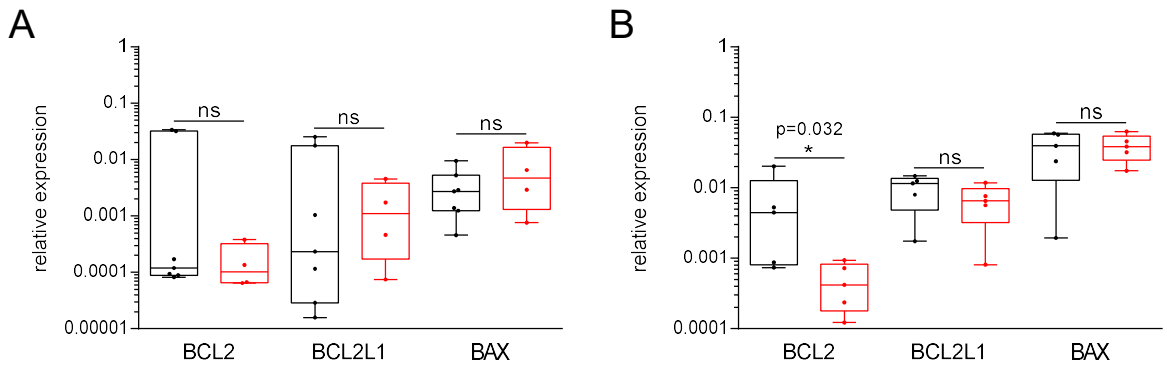

**Supplementary Figure S5.**

**Western blot analysis of p62/SQSTM1.**
A) UCi and (B) CPLC were subjected to Western blot analysis using autophagy-related marker p62/SQSTM1. One typical example is shown. Bar graphs depict relative expression in UCi (n=12) and CLPC (n=4) following densitometry analyses. GAPDH served for normalization. Mann-Whitney U test was used to test for statistical significance. ns, not significant $p>0.05$.




**Supplementary Figure S6.**

**CPLC are resistant to oxidative stress.** (A) Flow cytometry analysis of UCi and CLPC (B) following staining with Annexin V (AV) and 7AAD (AD). Wildtype cells were exposed to H202 for indicated time.




**Supplementary Figure S7.**

**qRT-PCR analysis of apoptosis-related genes.** Relative expression was determined in (A) UCi and (B) and CPLC. Box plots depicting expression in wildtype (black; n=7) and R236C cells (red; n=4) are shown. RT-qPCR data were normalized to GAPDH. Mann-Whitney U test was used to test for statistical significance. ns, not significant p>0.05.

**Supplementary table S1: Primers**

| Name | Gene | Sequence (left/right) | origin |
|---|---|---|---|
| *AFP* | Alpha fetoprotein | CAGGAAGTCTGCTTTGCTGAAG/ TCACACCGAATGAAAGACTCG | NM_001134.3 |
| *ALB* | Albumin | AGAAGTGCTGCAAGGCTGAC/ CCTAAGGCAGCTTGACTTGC | NM_000477 |
| *ATP7B* | ATPase, Cu++ transporting, beta polypeptide | TCCTCTGTGTCTGTGGTGCTC/ ATGCGCCTGTGCCTCATAC | NM_000053 |
| *BAX* | BCL2 Associated X | GCCCTTTTCTACTTTGCCAGC/ TCAGCCCATCTTCTTCCAGAT | NM_001291428.2 |
| *BCL2* | BCL2 Apoptosis Regulator | GGCCTTCTTTGAGTTCGGTGG/ GATAGGCACCCAGGGTGATGC | NM_000633.3 |
| *BCL2L1* | BCL2 Like 1 | TAAGGCGGATTTGAATCTC/ ATAATAGGGATGGGCTCAAC | NM_138578.3 |
| *CFTR* | CF transmembrane conductance regulator | GAAAGTTGCAGATGAGGTTGGGC/ TGCTTGTGGCCATGGCTTAGG | NM_000492.4 |
| *CK19* | Keratin 19 | AGGAGATTGCCACCTACCG/ AGAGGACCTTGGAGGCAGAC | NM_002276 |
| *CK7* | Keratin 7 | TGGGAGCCGTGAATATCTCTG/ GCACTGCTGGAGAAGCTCAG | NM_005556 |
| *CLDN1* | Claudin 1 | CCCAGTCAATGCCAGGTACG/ AAGTAGGGCACCTCCCAGAAG | NM_021101 |
| *FN1* | Fibronectin 1 | CTGGCCAGTCCTACAACCAG/ TCGGGAATCTTCTCTGTCAGC | NM_001306129 |
| *GAPDH* | Glyceraldehyde-3-phosphate oxygenase | CCCACTCCTCCACCTTTGAC/ CCACCACCCTGTTGCTGTAG | NM_002046 |
| *L1CAM* | L1 cell adhesion molecule | AGGGCGGCAAATACTCAGTG/ CGCCGAAGGTCTCATCTTTC | NM_000425 |
| *NANOG* | Nanog homeobox | AATGGTGTGACGCAGAAGG/ CTGGGGTAGGTAGGTGCTGA | NM_024865 |
| *OCLN* | Nuclear receptor subfamily 1 group I member 3 (*CAR*) | GAAGCAAGTGAAGGGATCTGC/ CCAACCATCTTCTTGATGTGTG | NM_001205254 |
| *OCT4* | POU class 5 homeobox 1 | GAACCGAGTGAGAGGCAACC/ AACCACACTCGGACCACATC | NM_002701 |
| *SNAI2* | Snail family zinc finger 2 | ACCCTGGTTGCTTCAAGGAC/ GAATGGGTCTGCAGATGAGC | NM_003068 |
| *SOX2* | SRY (sex determining region Y)-box 2 | ATGGGTTCGGTGGTCAAGTC/ CTGATCATGTCCCGGAGGTC | NM_003106 |
| *SOX9* | SRY (sex determining region Y)-box 9 | CCCAACGCCATCTTCAAGGC/ CCTGGGATTGCCCCGAGTG | NM_000346.4 |
| *TF* | Transferrin | GATAAGGAAGCTTGCGTCCAC/ TTGCCCGAGCAGTCAGTTAC | NM_001063 |
| *TTR* | Transthyretin | GAAAGGCTGCTGATGACACC/ TCAGTTGTGAGCCCATGCAG | NM_000371 |

**Supplementary table S2: Antibodies**

| Antibody | Method* | Company | Dilution | Catalog Number |
|---|---|---|---|---|
| AFP | ICC | Santa Cruz | 1:100 | sc-8399 |
| ALB | ICC | abcam | 1:100 | ab2406 |
| HNF4 | ICC | Santa Cruz | 1:100 | sc-6556 |
| NANOG | ICC | Stemcell Technologies | 1:100 | sc-33759 |
| OCT3/4-PE | ICC | Stemcell Technologies | 1:100 | 60093PE |
| SSEA-4-PE | ICC | Stemcell Technologies | 1:100 | 60062PE |
| Alexa Fluor 488 | ICC | Life Technologies | 1:500 | A11001 |
| Alexa Fluor 488 | ICC | Invitrogen | 1:500 | A11054 |
| Alexa Fluor 568 | ICC | Life Technolgies | 1:500 | A11057 |
| Alexa Fluor 594 | ICC | Life Technolgies | 1:500 | A11012 |
| LC3B | WB | Cell Signaling | 1:500 | 2775 |
| GAPDH | WB | Cell Signaling | 1:7000 | 2118 |
| p62/SQSTM1 | WB | Cell Signaling | 1:500 | 5114 |
| Anti-IgG | WB | Cell Signaling | 1:1000 | 7074 |
| CD13-PC5 | FC | Beckman Coulter | 1:100 | A07763 |
| CD29-FITC | FC | Beckman Coulter | 1:100 | IM0791U |
| CD71-FICT | FC | Beckman Coulter | 1:100 | IM0483 |
| CD105-PE | FC | Beckman Coulter | 1:100 | A07414 |
| CD166-PE | FC | Beckman Coulter | 1:100 | A22361 |
| AFP-PE | FC | Santa Cruz | 1:100 | sc-8399 PE |
| CK19-AF647 | FC | Santa Cruz | 1:100 | sc-6278 AF647 |
| SOX9-FITC | FC | Santa Cruz | 1:100 | sc-166505 FITC |
| FITC, PE, PC5 isotype control | FC | Beckman Coulter | 1:100 | A22361 |
| Alexa Fluor 647 isotype control | FC | Bio Legend | 1:100 | 366913 |

*ICC, immunocytochemistry; WB, Western blot; FC, flow cytometry