Supplementary Data

UASR1 Exon 1

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UASR1       1    CCCTCCTCAAACACACATCCATCCTCCGGcacacacccagtccatgcctcgcccccacac
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 5164 CCCTCCTCAAACACACATCCATCCTCCGGCACACACCCAGTCCATGCCTCGCCCCCACAC

UASR1       61   acacctgacaccccagtgcaacaccacacccaccctcccctccctgcaaactcccacctc
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 5104 ACACCTGACACCCCAGTGCAACACCACACCCACCCTCCCCTCCCTGCAAACTCCCACCTC

UASR1       121  CGCCCACCTGCTTAATACACATTCTCACCCCCCACACACTCCTTAATACATACTCTCACA
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 5044 CGCCCACCTGCTTAATACACATTCTCACCCCCCACACACTCCTTAATACATACTCTCACA

UASR1       181  CCCACAAGCCTGCCTTCTTGGAGAAGTGAGCCGAGCCGTGCAGCGCCGCGAAGG
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 4984 CCCACAAGCCTGCCTTCTTGGAGAAGTGAGCCGAGCCGTGCAGCGCCGCGAAGG
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UASR1 Exon 2

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UASR1       235  GCATCCCCGAAGACCGGGAGGAACGCCGCGGGGACCTGTGGCTTAGCGCGCTCCGCCCGG
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 4572 GCATCCCCGAAGACCGGGAGGAACGCCGCGGGGACCTGTGGCTTAGCGCGCTCCGCCCGG

UASR1       295  GCTTGTCTGCCCGCGGGGGCGCAGCGGCTGAGGCGGCTCCGGGCCGGAGTTCCAATCAAG
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 4512 GCTTGTCTGCCCGCGGGGGCGCAGCGGCTGAGGCGGCTCCGGGCCGGAGTTCCAATCAAG

UASR1       355  CGCCACCCAACTCCCAGTCGGGGGCCGAGGCCAGCGCCGGGATGCCAGCTTCCCCCAAAA
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 4452 CGCCACCCAACTCCCAGTCGGGGGCCGAGGCCAGCGCCGGGATGCCAGCTTCCCCCAAAA

UASR1       415  AGATCCTGCCTCAGGGAAATGCATGGAGCCGGCGGAAAAGCCCGCGGCGCCCCCGGCGGA
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 4392 AGATCCTGCCTCAGGGAAATGCATGGAGCCGGCGGAAAAGCCCGCGGCGCCCCCGGCGGA

UASR1       475  TCGCAGACCCTAAGGGGGCGGGAGGTGGCGCCCCAGTCCCAACCTCTTGAGCCAACCCAG
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 4332 TCGCAGACCCTAAGGGGGCGGGAGGTGGCGCCCCAGTCCCAACCTCTTGAGCCAACCCAG

UASR1       535  TGGGTGGGAAGTGCCCTTACCCTAGGCCTTCCGCAAAGTGTTCTCTCCTTGTATTATTCT
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 4272 TGGGTGGGAAGTGCCCTTACCCTAGGCCTTCCGCAAAGTGTTCTCTCCTTGTATTATTCT

UASR1       595  AATTACGGTATTTTTAATTTCCTTaaaaaaataagaaacagaaaagcacagaa
                 ||||||||||||||||||||||||||||||||||||||||||||||||||||
UNC5B-Int1 4212 AATTACGGTATTTTTAATTTCCTTAAAAAAATAAGAAACAGAAAAGCACAGAA
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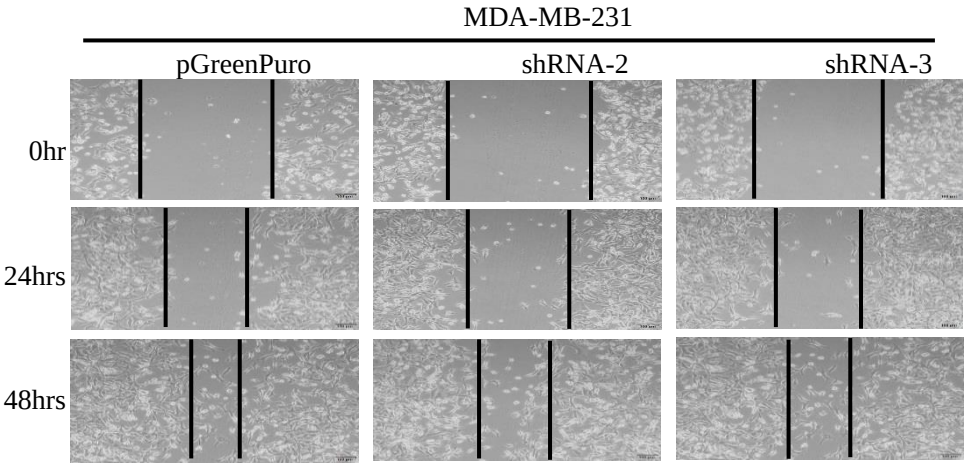
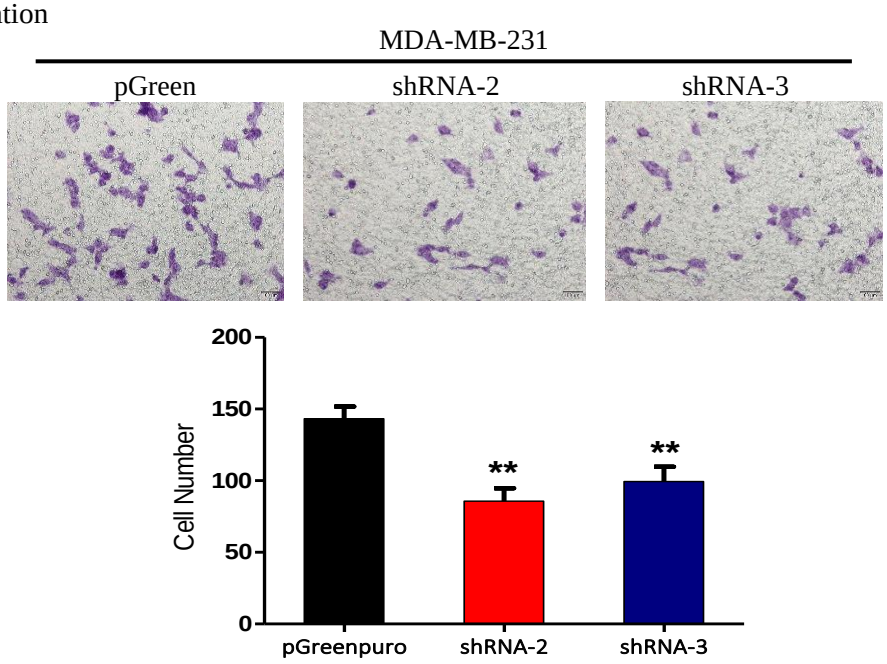
Supplementary Figure S1. Structure of UASR1 gene. Data show the blast between UASR1 cDNA and Intron 1 DNA sequence of UNC5B gene. UASR1 gene consists of two exons with 647bp in length and one intron of 358bp. UNC5B-Int1 indicates intron 1 of UNC5B gene, which is composed of 66,758bp.

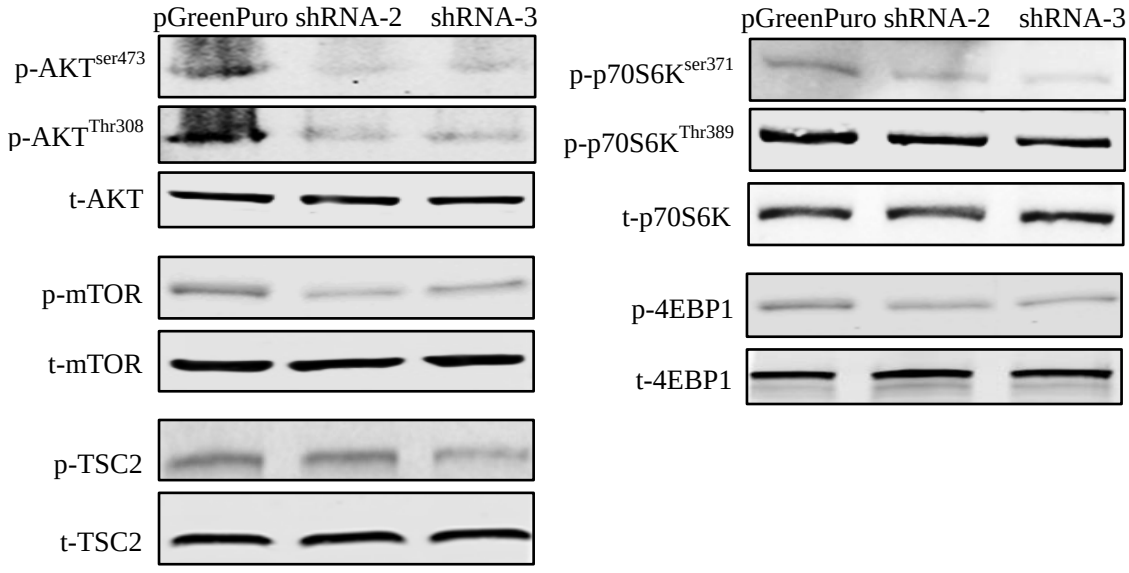
A) Wound healing



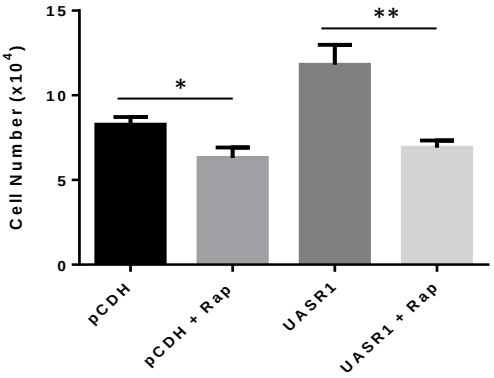
B) Migration



C) Western blot



Supplementary Figure S2. Effects of UASR1 silencing in MDA-MB-231 cells. UASR1 shRNAs were delivered by lentiviral vectors into MDA-MB-231 cells; would healing, migration assays and Western blot were conducted as described in Materials and Methods. (A) Wound healing. (B) Migration. Bar chart indicates cell number migrated. Data indicate mean ± SD from three independent experiments. ** $p<0.01$ compared to control. (C) Western blot, showing decrease of pAKT, p-mTOR, p-TSC2, p-p70S6K (Ser371 and Thr389), and p-4EBP1 proteins, i.e., inhibition of the AKT/mTOR pathway by UASR1 silencing.



Supplementary Figure S3. Rapamycin inhibits cell proliferation. MCF7 cells ($3x10^4$/well) were spread into 12-well plates. After incubation overnight, cells were fed with fresh medium containing rapamycin (20 nM) for 24 hours and then collected by trypsinization. After trypan blue staining, viable cells were counted using a Vi-cell counter. Rap indicates rapamycin. * $p<0.05$ and ** $p<0.001$.