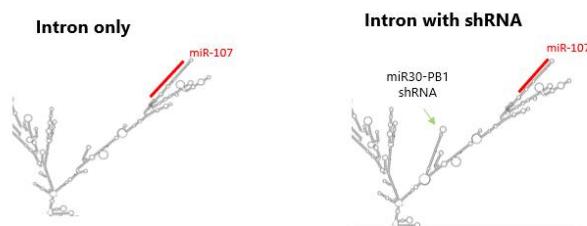


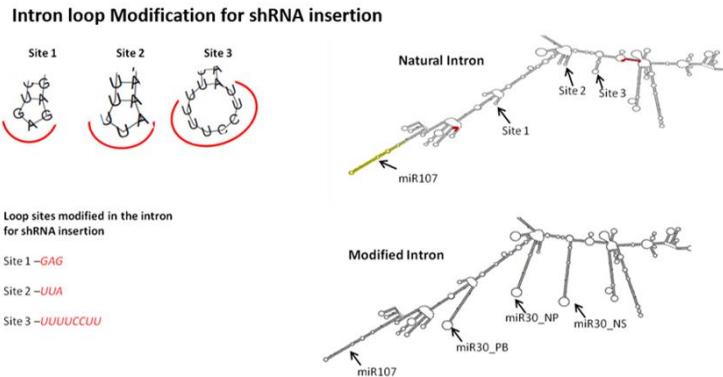
## Supplementary figure legends



**Figure S1.** Mfold schematics of shRNAs with traditional Brummelkamp (BK) loop and miR30 adapted structures. The PB1 siRNA sequence was embedded in the miR30 structure (miR30-PB1) with both the sense and antisense sequence connected by a loop. The natural miRNA sequence is shown alongside of miR30-PB1.



**Figure S2.** The predicted RNA secondary structures of a section of the natural intron 5 of PANK1 without (left) and with shRNA (right). The miR107 sequence is highlighted in red colour and the green arrow indicates the inserted miR30-PB1 shRNA within the intron. .



**Figure S3.** The selected loop sequences downstream of miR107 to be replaced with the miR30-PB1, miR30-NP and miR30-NS are highlighted in red colour. The comparison of the predicted secondary structure of the natural intron (top) and intron with three shRNAs (bottom) is also shown.

**Selection of three sites for shRNA insertion within intron 5:**

```
GGATCCATAGCATTGAGGACGGTAGTTGATGTTAAAGTACGCTTCACATCCTAGACAACATCTGAGAGATG
AATTATTTGGCTTAACAAAAGAGAACTTAATCAGTGCTTACATGTTTTTAAAAAAGGCTCAAATGTAG
CATATGTTGCAAGGCAGCTCTCTAGATAGTGTATTTTTCCTTAAATAAAAGCTATGTCTGATAAAACTCCAG
ACACCTACATTGTTTGAGGTTACC
```

Site 1  
Site 3  
Site 2

**Modified intron 5 with three shRNAs:**

```
GGATCCATAGCATTGAGGACGGTAGTTGATGTTAAAGTACGCTTCACATCCTAGACAACATCTGCGCGATC
TGTTCACTGCATGAAGACTGTGGAAGCAGCAGATGGGTCTCAATGGGAACAGATCTTGCAGATGAATTT
TTTGCTTAAACAAAAGAGAACTTAATCAGTGCTTACATGTTTTTGGAATCTTTTCTGCCGGAGACAAAC
TGGAAGCAGCAGATGGGTTGTCTCCGAAATAAGATTGCAAAAGGCTCAAATGTAGCATATTTTGCAAG
GCAGCTTCTAGATGTTTTGGAATGATACACAGTCCTGTGAAGCAGCAGATGGGGAA
CTGGTTTATCCATTCTGAAATAAGCTATGTTCTGATAAAACTCTCAGACCCTACATCTGTTTTTGAGGTTACC
```

Site 1 with PB1 shRNA  
Site 2 with NP shRNA  
Site 3 with NS shRNA

**Figure S4.** The selected intron 5 region of the PANK1 gene to insert three shRNAs. The three selected sites downstream to miR-107 are highlighted in red (top) and the intron sequence with PB1, NP and NS shRNA was provided (bottom). The intron containing three shRNAs (466 bp) was oligo-synthesised and used to clone into pGI vector.

**Table S1.** Synthesised oligonucleotides used in this study.

Name	sequence
GFP_fwd	ACTGTCGACATGGTGAGCAAGGGCGAGG
GFP_rwd	ACTGCGGCCGCTTACTTGTACAGCTCGTCC
SmaI_fwd	GACCCGGGCCGAGGTGAAG
SmaI_rev	CTTCACCTCGGCCCGGGTC
Intron_107_fwd	GTAAGATGGTGTATTACTATGAGC
Intron_107_rev	CTGTAAAATTGAAAAGCAG
BamHI_int_fwd	ACTAGATGGGATCCATAGC
BamHI_int_rev	GCTATGGATCCCCTAGT
KpnI_int_fwd	TGTGAGGTACCGCTCTAAC
KpnI_int_rev	GTTAGAGCGGTACCTCACAC

BK_PB1_fwd	GATCGATCTGTTCCACCATTGAATTCAAGAGATTCA AATGGTGGAACAGATCGTAC
BK_PB1_rev	GATCTGTCCACCATTGAATCTCTGAATTCAATGG TGGAACAGATC
miR30_ PB1_fwd	GATCGCGACAGATCTGTTCCACCATTGAAGACTGT GAAGCAGCAGATGGGCTTCAATGGAACAGAT CTGCTGCGTAC
miR30_PB1_rev	GCAGCAGATCTGTTCCCATTGAAGACCCCATCTGC TGCTTCACAGTCTCAATGGTGGAACAGATCTGTCG C
PB1_luc_fwd	CTCGAGGGAGATCATTGAGCTGAAG
PB1_luc_rev	GCGGCCGCCGTCTGAGTTCTCAATGG
NP_luc_fwd	GCGCTCGAGACCAAATGAGAATCCAGCAC
NP_luc_rev	AGCGCGGCCGCAATTGTCGTACTCCTCTGCA
NS_luc_fwd	ATGCTCGAGGTACTCTCGGTCTGGACATC
NS_luc_rev	ATCGCGGCCGTTCCGCCATTCTCGTTTC
miR-PTA	GAGGCGAGCACAGAATTAAATACGACTCCATATAG GTTTTTTTTTTVN
PAM-URP	GAGGCGAGCACAGAATTAAATACGAC
miR107	GCAGCAGCATTGTACAGGGCT
miR26a	GCCTCAAGTAATCCAGGATAGG
PB1	CGCTTCAATGGTGGAACAGATC
5S	TGGGAATACCGGGTGCTGT
Poly-A_fwd	ACTGCGGCCGCGACTCTAGATCA
Poly-A_rev	ACTGAATTACGCTTACAATTACGC

**Table S2.** siRNA sequences used in this study.

Name	Sequence (5' – 3')
PB1-2257	GATCTGTTCCACCATTGAA
NP	GATCTGTTCCACCATTGAA
NS1/NEP	AATGGAATGATAACACAGT