A

B BmCDK5-sgRNA/Cas9

```
WT     GCTTAAAAATCCTCTTTAATTGATCATCAACATCTGAACCTGGGAATAGCGGTCTGCCC
-7     GCTTAAAAATCCTCTTTAATT-------AACATCTGAACCTGGGAATAGCGGTCTGCCC
-9     GCTTAAAAATCCTCTTTAAT---------ACATCTGAACCTGGGAATAGCGGTCTGCCC
-12    GCTTAAAAATCCTCTTTAAT-----------TCTGAACCTGGGAATAGCGGTCTGCCC
-5     GCTTAAAAATCCTCTTTA-----TCATCAACATCTGAACCTGGGAATAGCGGTCTGCCC
-5/+1  GCTTAAAAATCCTCTTTA--C--TCATCAACATCTGAACCTGGGAATAGCGGTCTGCCC
```

C BmCNN-sgRNA/Cas9

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WT     AGCATGAAGCAGTATGAAGAACAGTTAAACGGATTGCGGAAGGAGAATTTTCACTTGA
-6     AGCATGAAGCAGTATG-----AGTTAAACGGATTGCGGAAGGAGAATTTTCACTTGA
-11    AGCATGAAGCAGTATGAAGAACAGTTAAA-------------AGGAGAATTTTCACTTGA
-8/+1  AGCATGAAGCAGTATGAAGAACAGTTAAA----A---GGAAGGAGAATTTTCACTTGA
-9     AGCATGAAGCAGTATGAAGAACAG----------TTGCGGAAGGAGAATTTTCACTTGA
-5/+1  AGCATGAAGCAGTATGAAGAACAG--G--CGGATTGCGGAAGGAGAATTTTCACTTGA
```

D

Phylogenetic tree:
- centrosomin-like isoform X2 *Bombus terrestris*
- centrosomin-like isoform X1 *Bombus terrestris*
- centrosomin isoform X3 *Bombus impatiens*
- centrosomin isoform X2 *Bombus impatiens*
- centrosomin isoform X1 *Bombus impatiens*
- CDK5 regulatory subunit-associated protein 2 isoform X2 *Apis florea*
- intracellular protein transport protein USO1 isoform X3 *Apis mellifera*
- intracellular protein transport protein USO1 isoform X2 *Apis mellifera*
- intracellular protein transport protein USO1 isoform X1 *Apis melliferahoney bee*
- centrosomin isoform X5 *Megachile rotundata*
- centrosomin isoform X1 *Megachile rotundata*
- CDK5 regulatory subunit-associated protein *Cerapachys biroi*
- CDK5 regulatory subunit-associated protein 2 isoform X3 *Athalia rosae*
- centrosomin isoform X1 *Tribolium castaneumred flour beetle*
- centrosomin isoform X2 *Tribolium castaneumred flour beetle*
- centrosomin isoform A *Drosophila melanogaster*
- centrosomin isoform B *Drosophila melanogaster*
- GJ21311 *Drosophila virilis*
- centrosomin isoform X2 *Ceratitis capitata Mediterranean fruit fly*
- centrosomin isoform X3 *Bactrocera cucurbitae*
- centrosomin isoform X1 *Bactrocera cucurbitae*
- centrosomin isoform X2 *Bactrocera cucurbitae*
- CDK5 regulatory subunit-associated protein 2 *Zootermopsis nevadensis*
- hypothetical protein KGM 02049 *Danaus plexippus*
- ● centrosomin isoform X2 *Bombyx mori*
- ● centrosomin isoform X1 *Bombyx mori*
- CDK5 regulatory subunit-associated protein 2-like isoform X2 *Plutella xylostella*
- CDK5 regulatory subunit-associated protein 2-like isoform X1 *Plutella xylostella*
- calponin-3 isoform 1 *Homo sapiens*
- calponin-3 isoform 2 *Homo sapienshuman*
- calponin-3 isoform 3 *Homo sapiens*

Figure. S1. (A) The level of transcription of *BmCDK5 and BmCNN* in *BmCDK5* overexpressing BmNS cells and empty vector control cells was determined using qRT-PCR. The level of transcription of *BmCNN* in *BmCNN* overexpressing BmNS cells and empty vector control cells was determined using qRT-PCR (* $p < 0.05$, ** $p < 0.01$).

(B-C) DNA sequencing analysis of the CRISPR/Cas9 editing target genes (BmCDK5 and BmCNN). The red sequence is the target sequence. WT is the unknocked sample, as a reference. (D) Phylogenetic tree of CNN.

Table S1. The RT-qPCR primer sequences.

Primers	Sequences (5'-3')
QBmCDK5/F	GATGTATTGCACAGCGAAAAGA
QBmCDK5/R	CTTCAGCGGAATAGCACTTTAC
QBmCNN/F	GCACAACGTTCGATTACAAGTA
QBmCNN/R	TTTCATCGAAGCGCTATTGTTC
BmCDK5/F	CGCGGATCCATGGATTACAAGGATGACGACGATAAGATGCAAAAATAT
BmCDK5/R	CCGGAATTCTTAACATCGATCATTCTTGACTG

1

BmCNN/F	CCGGAATTCATGTACCCATACGATGTTCCAGATTACGCTATGGCCACATTACCTAGAAC
BmCNN/R	CCGCTCGAGTTACATATTACTATGAGTTTCG
sgBmCDK5/F	AAGTGATCATCAACATCTGAACCT
sgBmCDK5/R	AAACAGGTTCAGATGTTGATGATC
sgBmCNN/F	AAGTACAGTTAAACGGATTGCGGA
sgBmCNN/R	AAACTCCGCAATCCGTTTAACTGT
Qsw22934/F	AACACCCCGTCCTGCTCACTG
Qsw22934/R	GGGCGAGACGTGTGATTTCCT
M13/F	GTTTTCCCAGTCACGAC
M13/R	CAGGAAACAGCTATGAC
U6/F	GGAAGATCTTCCAGGTTATGTAGTACACATTGT
U6/R	GGAAGATCTTCCAAAAAAAGCACCGACTCG
T-BmCDK5/F	CGCGGATCCATGCAAAAATATGAG
T-BmCDK5/R	GGAGATTTATTATGATGAACTCACC
T-BmCNN/F	TACTGGTGGTAGTACTTAGA
T-BmCNN/R	ATATCGATTTTATCAAGCAC