## **Supplementary Material**





**Fig. S1.** Subcellular location of CEP2 and MyHC in myoblasts and myotubes after transfection. (A) Subcellular location of CEP2 exogenous protein in myoblasts. Recombinant plasmids were transfected into C2C12 cells using lipofectamine 2000. The fluorescent signals were analyzed by confocal microscopy. The arrows indicate C2C12 cells (B) Subcellular location of CEP2 and MyHC in myotubes. The nuclei were stained blue with DAPI. MyHC was detected by anti-myosin (skeletal, fast) antibody (green). CEP2 was probed with anti-CEP2 antibody (red). Scale bar =  $25\mu$ m. n = 3. The arrows indicate one myotube.

## **Figure S2**



**Fig. S2.** Up-regulation of *CEP2* mediated by lentivirus and its effect on myogenesis. (A) Lentivirus-mediated overexpression of *CEP2* was determined in GM on the  $2^{nd}$  d and in DM on the  $10^{th}$  d, respectively. (B) qPCR analysis of *CEP2* both in GM on the  $2^{nd}$  d and in DM on the  $10^{th}$  d. \*p<0.05;  $n \ge 3$ . (C) Subcellular location of CEP2-GFP protein in myoblasts and myotubes, respectively. Scale bar =  $25\mu$ m. The arrows indicate C2C12 cells. (D) RNA detection of myogenic factors in DM on the  $10^{th}$  d. \*p<0.05;  $n \ge 3$ . (E) Western blotting analysis of myogenic proteins showed different change in DM on the  $10^{th}$  d. (F) Protein level of myogenic factors are presented as mean  $\pm$  S.E.M. \*p<0.05;  $n \ge 3$ . (G) Subcellular location of CEP2-GFP and MyHC in a myotube. MyHC was detected with anti-myosin (skeletal, fast) antibody (red). Scale bar =  $25\mu$ m. The arrows indicate one myotube.

Figure S3.



**Fig. S3.** Knockdown of *CEP2* did not influence cell proliferation. (A) The cells were fixed for EDU (red) immunostaining after transfection for 24 hours. Scale bar = 50 µm. (B) The proportion of proliferation cells in EDU assay was presented as mean  $\pm$  S.E.M (n=3). (C) C2C12 cells were collected for PI staining and FACSCalibur cell cycle assay. (D) The proliferative index in FACSCalibur assay was presented as mean  $\pm$  S.E.M (n=3). (E) qPCR analysis of cell cycle-specific genes at 24 h post-transfection. siCEP2: siRNA pool targeted to *CEP2*, NC: native control (No target siRNA pool), siLamin A: positive control (siRNA pool targeted to *Lamin A*). \**p*<0.05; n ≥ 3.

SMARTpool contentSequenceTarget Sequence 1GACCUUCCCUUCCAGUUUATarget Sequence 2GAUUAUGGAUCACGACCUATarget Sequence 3UGGCGGAGAUGACAUGUUUTarget Sequence 4CGUGCAGAUUCCUACAUAA

Table S1. siGENOME Mouse Cdc42ep2 (104252) siRNA-SMARTpool, 5 nmol

Tabl	e S2.	Primers	used	for	qPCR
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Name	Sequence
CEP2	S: 5' TCCCCATCTATTTGAAACGTGG 3'
	A: 5' CCGCTGTTCCTGGAAGGAG 3'
Lamin A	S: 5' CCACCGAAGTTCACCCTAAA 3'
	A: 5' CTCGTCGTCATCCTCATTGTC 3'
P21	S: 5' CCTGGTGATGTCCGACCTG 3'
	A: 5' CCATGAGCGCATCGCAATC 3'
cyclin D1	S: 5' GCGTACCCTGACACCAATCTC 3'
	A: 5'ACTTGAAGTAAGATACGGAGGGC 3'
Cdk4	S: 5' ATGGCTGCCACTCGATATGAA 3'
	A: 5' TCCTCCATTAGGAACTCTCACAC 3'
cyclin E	S: 5' ATGTCAAGACGCAGCCGTTTA 3'
	A: 5' GCTGATTCCTCCAGACAGTACA 3'
myf5	S: 5' CCTGTCTGGTCCCGAAAGAAC 3'
	A: 5' GACGTGATCCGATCCACAATG 3'
myoD	S: 5' GCCTGAGCAAAGTGAATGAG 3'
	A: 5' GCAGACCTTCGATGTAGCG 3'
myogenin	S: 5' GCAATGCACTGGAGTTCG 3'
	A: 5' ACGATGGACGTAAGGGAGTG 3'
mrf4	S: 5' CTACATTGAGCGTCTACAGGACC 3'
	A: 5' CTGAAGACTGCTGGAGGCTG 3'
troponin T2	S: 5' CAGCAGCGTATTCGCAATGA 3'
	A: 5' TCTGGATGTACCCTCCAAAGTG 3'
MyHC 2b	S: 5'AGCTTGAAAACGAGGTGGAA 3'
	A:5' CCTCCTCAGCCTGTCTCTTG 3'
$\beta$ -actin	S: 5' TGTTACCAACTGGGACGACA 3'
	A:5' CTGGGTCATCTTTTCACGGT 3'