Supplementary Figure S1. Ectopic NEDD9 expression partially reverses the inhibitory function of LKB1 upon cell migration and invasion of human lung cancer cells. A, the wound healing kinetics of A549 cells with ectopic LKB1 and/or NEDD9 expression. Data were shown as mean ± s.e.m.. B and C, invasivion of A549 cells with ectopic LKB1 and/or NEDD9 expression were assessed by boyden chamber assay. Representative photos (B) and the migrated cell number per high-power field (HPF, C) were shown. Scale bar: 50μm (B). Data were shown as mean ± s.e.m.. *** $P < 0.001$.

Supplementary Figure S2. sh*Nedd9* inhibits lung cancer progression in *de novo* $Kras^{G12D}$, $Lkb1^{L/L}$ mouse model. A and B, real-time RT-PCR (A) and immunostaining (B) analyses confirmed the knockdown of *Nedd9* in lung tumors from $Kras^{G12D}$, $Lkb1^{L/L}$ mice at 21 weeks post virally infected with either Ctrl-*Cre* or sh*Nedd9*-*Cre* (7 mice per group). Data were shown as mean ± s.e.m.. * $P < 0.05$ (n=6 for Ctrl-*Cre* group and n=7 for sh*Nedd9*-*Cre* group) (A). Scale bar: 50μm (B). C, quantification of tumors in H&E-stained lung sections from $Kras^{G12D}$, $Lkb1^{L/L}$ mice virally infected with Ctrl-*Cre* or sh*Nedd9*-*Cre*. Data were shown as mean ± s.e.m.. $P > 0.05$. D, gross inspection showed the tumor nodules (indicted by yellow arrows) visible on lung surface from $Kras^{G12D}$, $Lkb1^{L/L}$ mice virally infected with Ctrl-*Cre* or sh*Nedd9*-*Cre*. E, quantification of tumor area in H&E-stained lung sections from $Kras^{G12D}$, $Lkb1^{L/L}$ mice virally infected with Ctrl-*Cre* or sh*Nedd9*-*Cre*. Data were shown as mean ± s.e.m.. $P > 0.05$. F, cleaved caspase-3 immunostaining (indicated by the red arrows)

on lung sections from *Kras*^{G12D}, *Lkb1*$^{L/L}$ mice virally infected with Ctrl-*Cre* or sh*Nedd9*-*Cre*. Scale bar: 50μm.

Supplementary Figure S3. NEDD9 promotes lung cancer progression in *de novo* *Kras*^{G12D} mouse model. A and B, real-time RT-PCR (A) and immunostaining (B) analyses confirmed the expression of NEDD9 in lung tumors from *Kras*^{G12D} mice at 24 weeks post virally infected with Lenti-*Cre* or *NEDD9*-*Cre* (5 mice per group). The primer recognizes both human and mouse *NEDD9*. Data were shown as mean ± s.e.m.. * $P < 0.05$ (n=5 for Lenti-*Cre* group and n=5 for *NEDD9*-*Cre* group) (A). Scale bar: 50μm (B). C, quantification of tumors in H&E-stained lung sections from *Kras*^{G12D} mice virally infected with Lenti-*Cre* or *NEDD9*-*Cre*. Data were shown as mean ± s.e.m.. $P > 0.05$. D, gross inspection of the tumor nodules (indicated by yellow arrows) visible on lung surface from *Kras*^{G12D} mice virally infected with Lenti-*Cre* or *NEDD9*-*Cre*. E, quantification of tumor area in H&E-stained lung sections from *Kras*^{G12D} mice virally infected with Lenti-*Cre* or *NEDD9*-*Cre*. Data were shown as mean ± s.e.m.. $P > 0.05$. F, cleaved caspase-3 immunostaining (indicated by the red arrows) on lung sections from *Kras*^{G12D} mice virally infected with Lenti-*Cre* or *NEDD9*-*Cre*. Scale bar: 50μm.

Supplementary Figure S4. LKB1 specifically regulates CRE-Luc activity. A, *LKB1* knockdown in CRL-5866 cells specifically up-regulated the activity of CRE but not other reporters as indicated. Data were shown as mean ± s.e.m.. ** $P < 0.01$. B, ectopic expression of LKB1 decreased the activity of CRE reporter but not that of

AP1 reporter in A549 cells. Data were shown as mean ± s.e.m.. ** $P < 0.01$.

Supplementary Figure S5. sh*CRTC1* down-regulates *NEDD9* mRNA expression. A, *CRTC1* knockdown efficiency in A549 cells was assessed by real-time RT-PCR quantification. Data were shown as mean ± s.e.m.. B, *CRTC1* knockdown in A549 cells down-regulated *NEDD9* mRNA level assessed by real-time RT-PCR. Data were shown as mean ± s.e.m.. * $P < 0.05$.

Supplementary Figure S6. Screening for the responsive element of CRTC1/CREB on *NEDD9* promoter. A, scheme of a series of *NEDD9* reporters with deletions. B, reporter gene assay for *NEDD9* promoters were performed in CRL-5866 cells with ectopic expression of CRTC1 and CREB. C and D, reporter gene assay were performed using *NEDD9* reporters with deletion mutations from -321bp to -115bp (C) or from -186bp to -146bp (D) of *NEDD9* promoter after ectopic expression of CRTC1 in CRL-5866 cells. Data were shown as mean ± s.e.m..

Supplementary Figure S7. LKB1 regulates *NEDD9* promoter activity through the non-classical CRE site. A, *LKB1* knockdown increased the transcription activity of *NEDD9* promoter with wild-type but not mutated non-classical CRE site in CRL-5866 cells. B, LKB1 down-regulated the transcription activity of *NEDD9* promoter with wild-type but not mutated non-classical CRE site in A549 cells. Data were shown as mean ± s.e.m..

Supplementary Figure S8. Immunofluorescence assay showed that ectopic Flag-LKB1 expression promoted SIK2 translocation into nucleus in A549 cells.

Flag-LKB1 (green), SIK2 (red), DAPI (blue). Scale bar: 10μm.

Supplementary Figure S9. Immunofluorescence assay showed that *LKB1* knockdown promoted SIK2 translocation into cytoplasm while CRTC1 translocation into nucleus in CRL-5866 cells. Scale bar: 10μm.

Supplementary Figure S10. *SIK2* knockdown up-regulates *NEDD9* mRNA expression via promoting CRTC1 nuclear translocation. A, *SIK2* knockdown efficiency in CRL-5907 cells was assessed by real-time RT-PCR quantification. Data were shown as mean ± s.e.m.. B, *SIK2* knockdown in CRL-5907 cells up-regulated *NEDD9* mRNA level assessed by real-time RT-PCR. Data were shown as mean ± s.e.m.. *** $P < 0.001$. C, immunofluorescence assay showed that *SIK2* knockdown promoted CRTC1 translocation into nucleus in CRL-5907 cells. Scale bar: 10μm.

Supplementary Figure S11. Ectopic CRTC1 expression partially reverses the inhibitory function of LKB1 upon cell migration and invasion of human lung cancer cells. A, the wound healing kinetics of A549 cells with ectopic LKB1 and/or CRTC1 expression. Data were shown as mean ± s.e.m.. B and C, invasion of A549 cells with ectopic LKB1 and/or CRTC1 expression were assessed by boyden chamber assay. Representative photos (B) and the migrated cell number per high-power field (HPF, C) were shown. Scale bar: 50μm (B). Data were shown as mean ± s.e.m.. *** $P < 0.001$.

Supplementary Figure S12. NEDD9 immunostaining in human lung cancer specimens. Representative photos were shown for tumors with low or high NEDD9 expression. Scale bar: 50μm.