Supplementary data

TRIM31 is up-regulated in hepatocellular carcinoma and promotes disease progression by inducing ubiquitination of TSC1-TSC2 complex

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Supplementary Figure 1. Verification of the specification of TRIM31 antibody. HepG2 and HuH7 cells were transfected with SiRNA specifically targeting TRIM31 (Si-TRIM31) or non-specific control (Si-NC). The cells were further cultured for 24h before being fixed and stained with TRIM31 antibody at 4°C overnight. The cells were then incubated with FITC conjugated secondary antibody at 37°C for 1 h and the nucleuses were stained with DAPI for 5 minutes. The immunofluorescence was investigated by immunofluorescence microscopy.
Supplementary Figure 2. Screening of signaling transduction pathways in HCC cells by TRIM31 overexpression. (a,b) BEL7402, SMMC7721 and HepG2 cells were transfected with TRIM31 expression plasmid or mock control, the activation of mTORC1 pathway (a) and MAPK pathway (b) were detected by western blot. (c,d) BEL7402 and HepG2 cells were transfected with TRIM31 expression plasmid or mock control, the activation of NF-κB pathway (c) and AMPK pathway (d) were detected by western blot.
Supplementary Figure 3. Screening of signaling pathway in TRIM31-knockdown cells. (a-c) BEL7402 and SMMC7721 cells were transfected with SiRNA specifically targeting TRIM31(Si-TRIM31) and non-specific control(Si-NC). The cells were further cultured for 24h and the activation of mTORC1 pathway(a), MAPK pathway(b) and NF-kappaB pathway(c) were detected by western blot.
Supplementary Figure 4. Detection of autophagy in HCC cells. (a) The SMMC7721 cells and BEL7402 cells were transfected with TRIM31 plasmid or mock control and further cultured for 24h, the levels of autophagy related proteins were detected by western blot. (b) SMMC7721 and BEL7402 cells were transfected with SiRNA specifically targeting TRIM31 (Si-TRIM31) and non-specific control (Si-NC) and further cultured for 24h. The levels of autophagy related proteins were detected by western blot.