Figure S1. Viability of NCI-H460, HEpG2 and 293 cells following treatment with 1 nM sufentanil for 24 h assessed by MTT assay. Sufen, sufentanil.

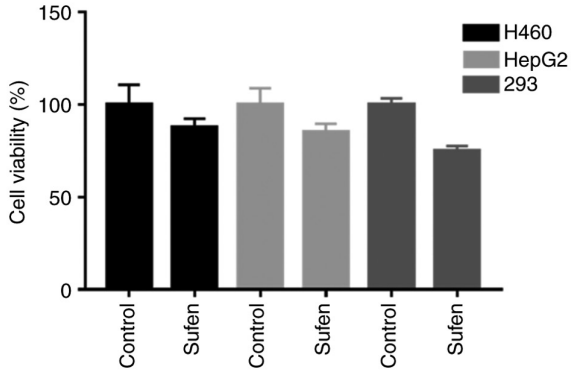

Figure S2. Western blot analysis of p62, LC3 and β-actin in 293 and HepG2 cells treated with 1 nM sufentanil or PBS for 24 h. Sufen, sufenanil.

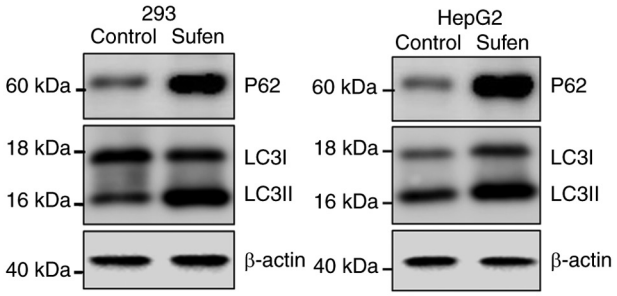

Figure S3. Western blotting analysis of Beclin1 and β-actin in NCI-H460 cells treated with 1 nM sufentanil or PBS for 0, 4 and 24 h.

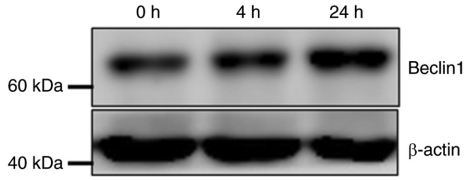

Figure S4. Western blot analysis of Beclin1, CathD, P62 and β-actin in NCI-H460 cells treated with 1 nM sufentanil or PBS with or without CQ for 24 h. CathD, cathepsin D; CQ, chloroquine; Sufen, sufentanil.

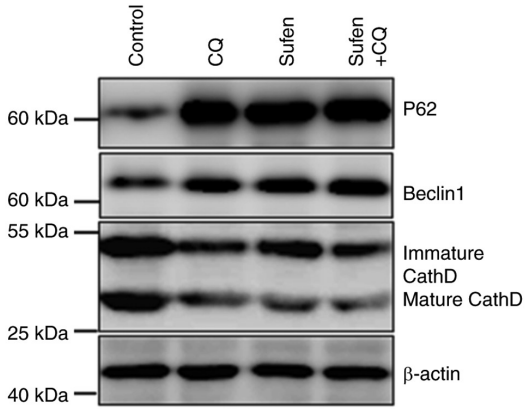

Figure S5. Autophagy was involved in the inhibition of migration by sufentanil in 293 and HepG2 cells. (A and B) Sufentanil suppressed wound closure. Scratched 293 and HepG2 cells were treated with 1 nM of sufentanil, PBS, 1 nM sufentanil + 50 μM CQ or 1 nM sufentanil + 100 mM trehalose for 24 h. Scale bar, 1 mm. Wound closure quantification from (A and B). (C and D) Cell invasion images. Scale bar, 50 μm. Quantification of the invaded cells per field. *P<0.05, **P<0.01 and ***P<0.001 (comparison between any two means). CQ, chloroquine; NS, not significant; Sufen, sufentanil; Tre, trehalose.