

Supplementary Information

Figure S1. Xylocyidine does not activate the caspases even at a concentration of 75 μM of treatment. HeLa cells were treated with 0.15% DMSO (v/v) (Control) or with 75 μM xylocyidine for the indicated times. (A–C) Cell-free caspase-3, -8 and -9 activities were measured. (D) Whole-cell lysates were analyzed by immunoblotting for caspase-8, caspase-9, PARP and β -actin.

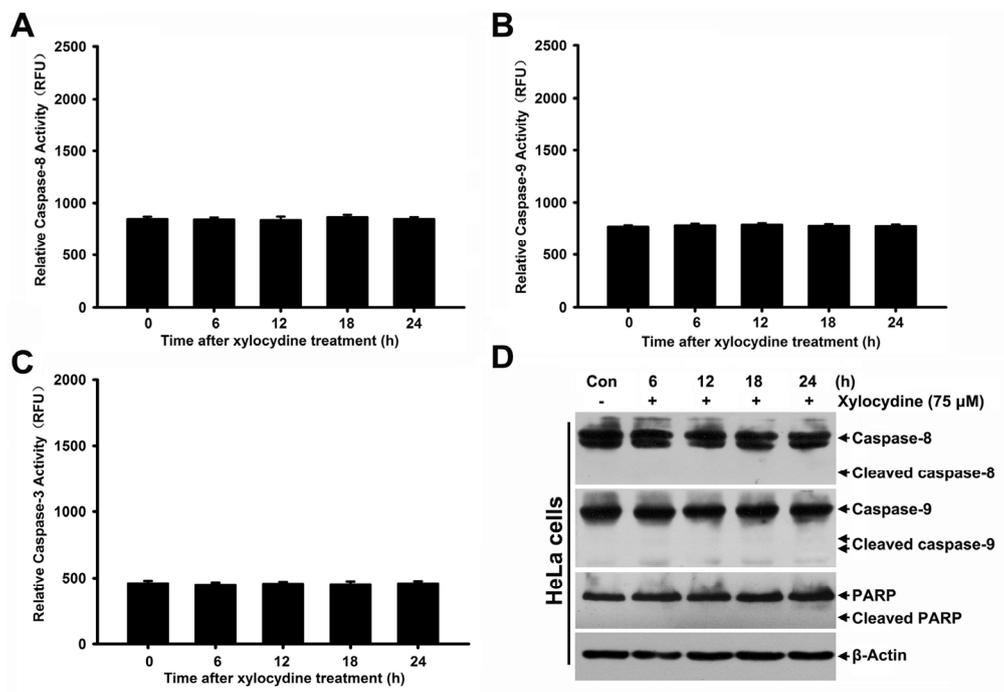


Figure S2. JRS-15 does not induce the cleavages of caspases and PARP in LO2 cells at a concentration of 30 μM of treatment. LO2 cells were treated with 0.1% DMSO (v/v) (Control) or with 30 μM JRS-15 for the indicated times. Whole-cell lysates were analyzed by immunoblotting for caspase-8, caspase-9, PARP and β -actin.

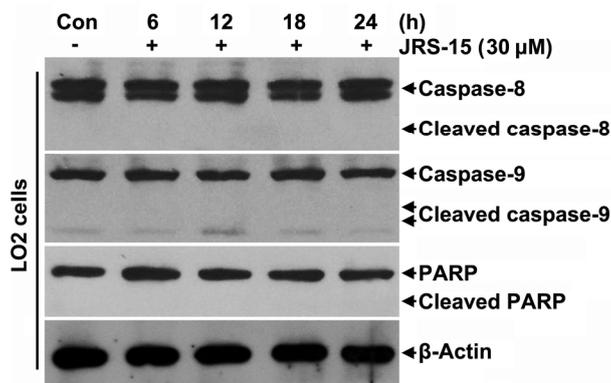
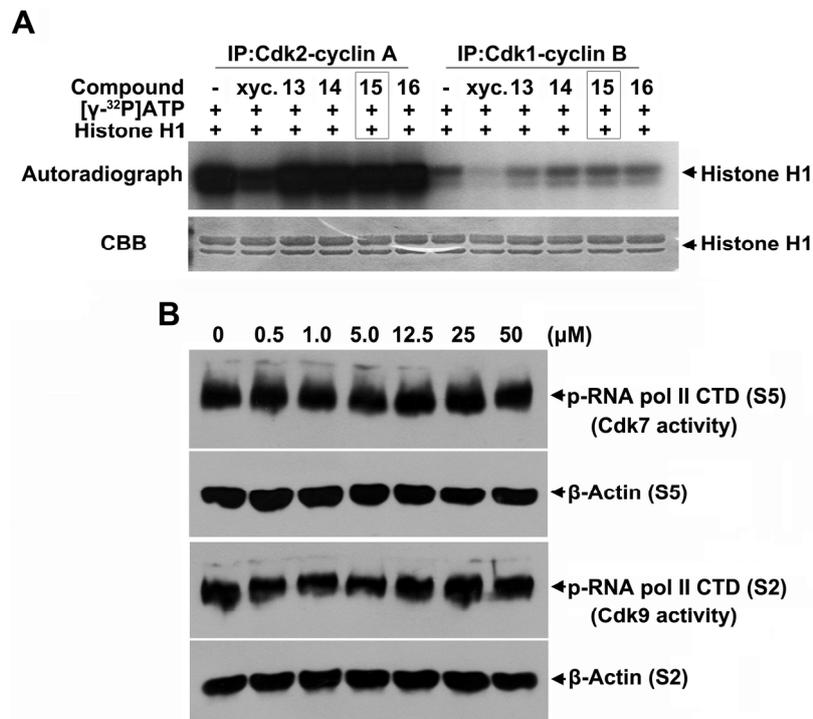


Figure S3. JRS-15 cannot inhibit Cdk1 and Cdk2 activity *in vitro*, as well as Cdk7 and Cdk9 activity in cultured HeLa cells. **(A)** The kinase assays were performed at 30 °C for 15 min in the reaction buffer (50 mM Tris-HCl (pH 7.4), 10 mM MgCl₂, 1 mM EGTA, 50 mM β-glycerophosphate, 0.1 mM Na₃VO₄, 25 mM NaF, 2 mM DTT, protease inhibitors and 0.1 mM phenylmethylsulfonyl fluoride) with a final volume of 50 μl that containing 5 μg Histone H1 protein as a specific substrate, Cdk1/cyclin B or Cdk2/cyclin A immunoprecipitated from protein extract of HeLa cells as an enzyme source, 10 μCi of [γ -³²P]ATP, 15 μM ATP and 10 μM compound. Samples were analyzed by 12% SDS-PAGE followed by autoradiography. The compound of number 15 represents JRS-15, and xylocyidine (xyc.) as a positive control. **(B)** HeLa cells were treated with the indicated concentrations of JRS-15 for 24 h. Whole-cell lysates were analyzed for Cdk7/9-specific phosphorylation of RNA polymerase II-CTD by immunoblotting, β-actin as a loading control.



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