

## Supplemental information

### Wip1 inhibitor GSK2830371 inhibits neuroblastoma growth by inducing Chk2/p53-mediated apoptosis

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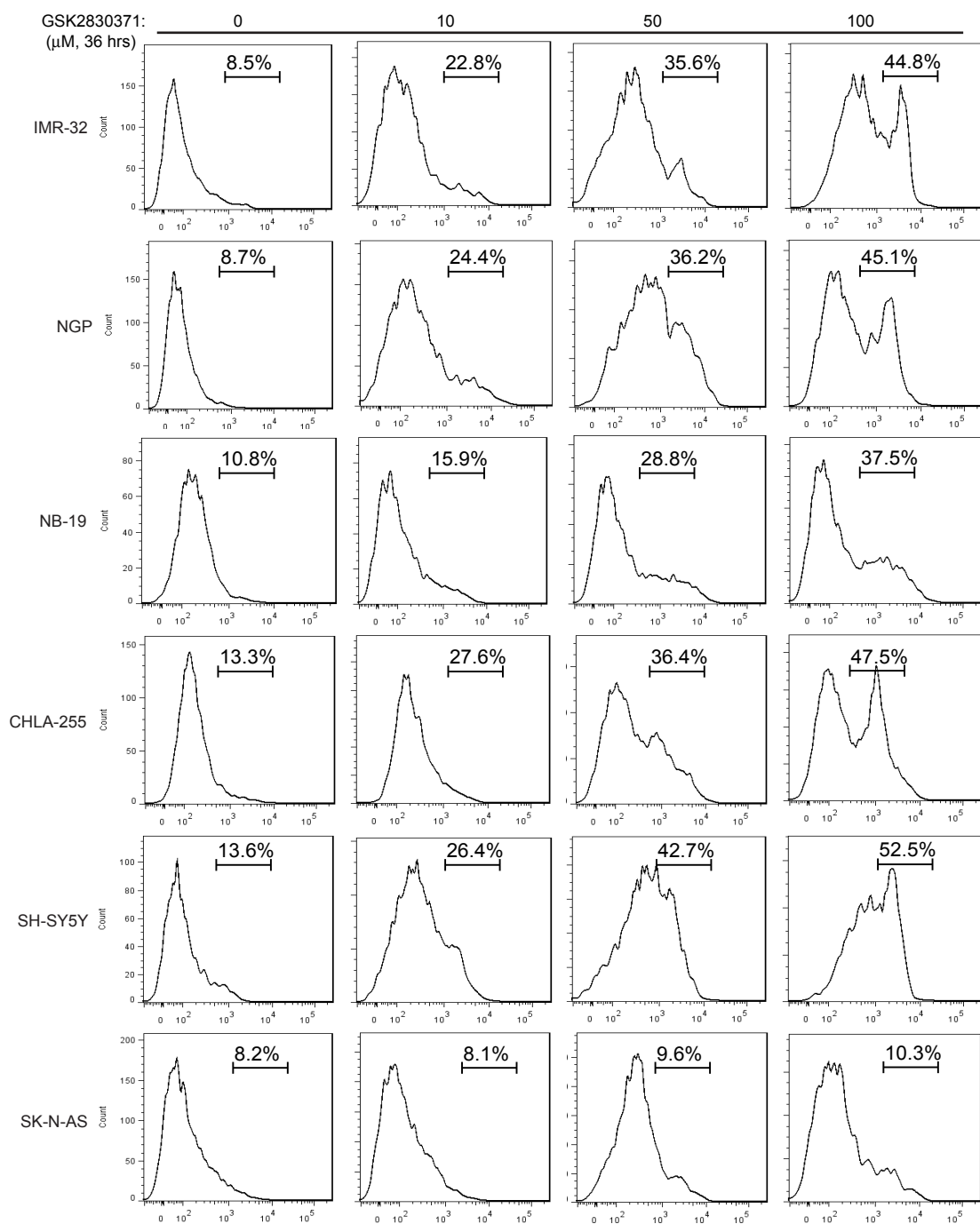
#### Running head: Wip1 inhibition in neuroblastoma

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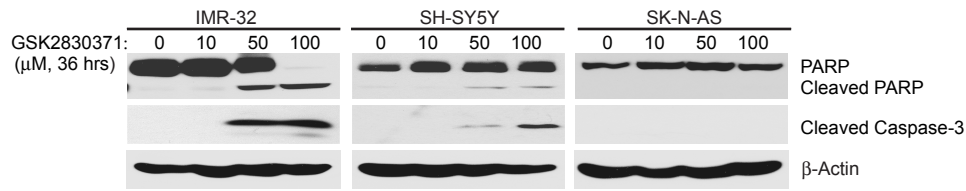
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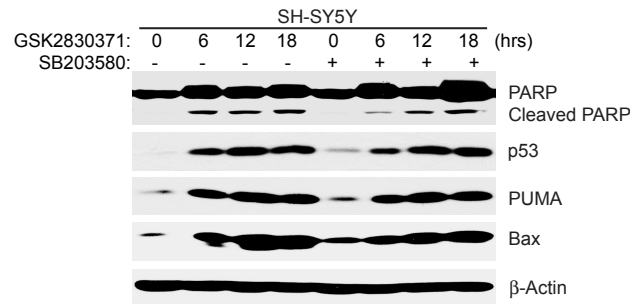
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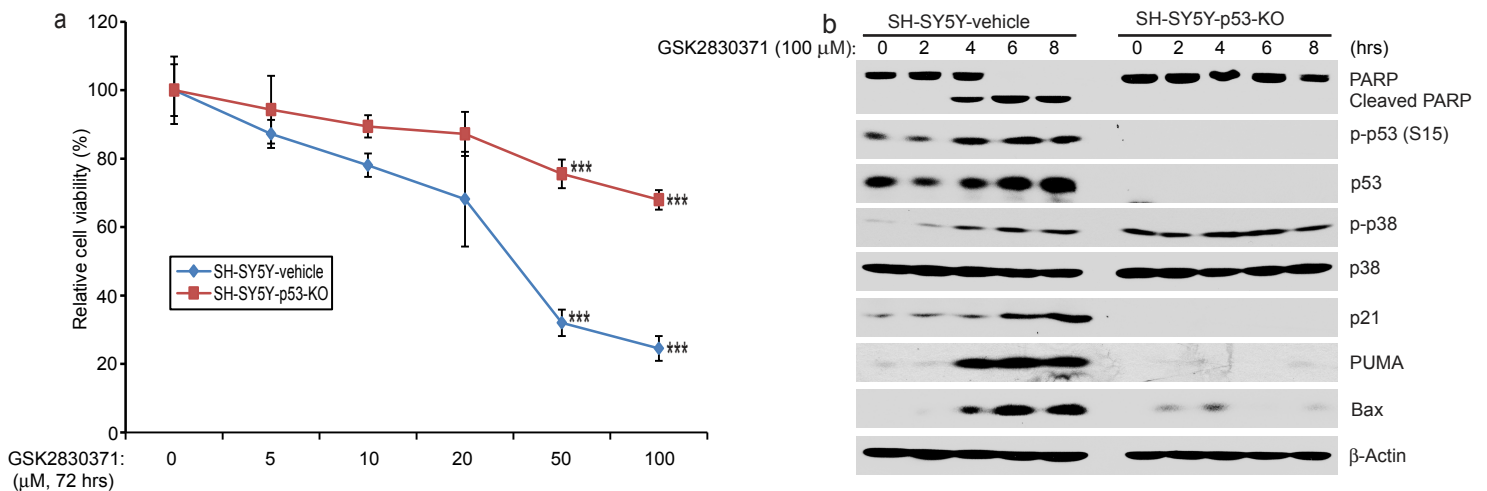
**Supplemental Figure S1. GSK2830371 induces cell death in *p53* wild-type, but not in *p53* mutant NB cells.** PI staining of IMR-32, NGP, NB-19, CHLA-255, SH-SY5Y, and SK-N-AS cells after treated with the indicated concentrations of GSK2830371 for 36 hrs. The cells were then analyzed by flow cytometry for the percentage of dead cells.



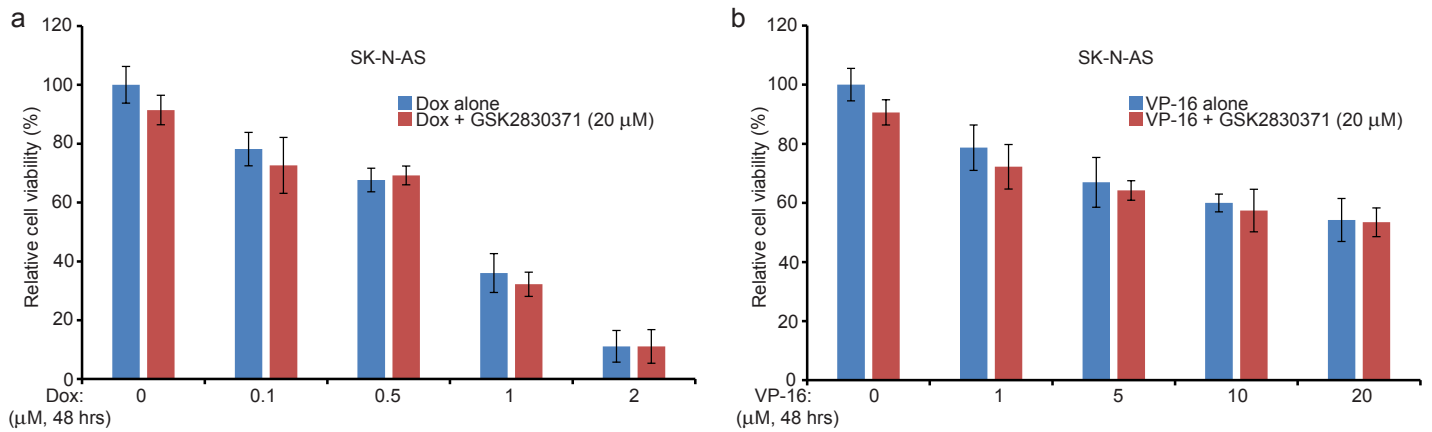
**Supplemental Figure S2. GSK2830371 induces apoptosis in *p53* wild-type, but not in *p53* mutant NB cells.** IMR-32, SH-SY5Y and SK-N-AS cells were treated with the indicated concentrations of GSK2830371 for 36 hrs. The cells were then lysed and subjected to SDS-PAGE followed by immunoblotting with anti-PARP and Caspase-3 antibodies.  $\beta$ -Actin was used as a loading control.



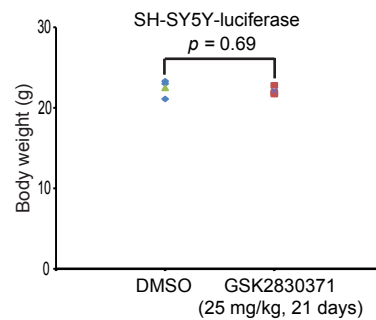
**Supplemental Figure S3. p38 inhibitor SB203580 suppresses GSK2830371-induced cell death and p53 activation.** SH-SY5Y cells were treated with or without p38 inhibitor SB203580 (10  $\mu$ M) for 2 hrs before exposed to GSK2830371 (50  $\mu$ M) for indicated time points (0-18 hrs). Cells were collected and lysed at the end of treatment followed by SDS-PAGE, and immunoblotting with the indicated antibodies.  $\beta$ -Actin was used as a loading control.



**Supplemental Figure S4. p53 activation plays a major role in GSK2830371-induced cytotoxicity in SH-SY5Y cells.** (a) SH-SY5Y-vehicle control cells and SH-SY5Y-*p53*-KO cells were seeded in 96-well plates, respectively. After incubating with the increasing concentrations of GSK2830371 for 72 hrs, cell viability was then measured by the CCK-8 assay and the results were represented as % vehicle  $\pm$  S.D.  $P < 0.001$  (\*\*\*) (Student's t-test, two-tailed) was indicated. (b) SH-SY5Y-control cells and SH-SY5Y-*p53*-KO cells were treated with GSK2830371 (100  $\mu$ M) for the indicated time points (0-8 hrs). And then the cells were harvested and lysed, followed by SDS-PAGE and immunoblotting with the indicated antibodies.  $\beta$ -Actin was used as a loading control.



**Supplemental Figure S5. GSK2830371 fails to enhance Dox- and VP-16-induced cytotoxicity in *p53* mutant SK-N-AS cells.** SK-N-AS cells were seeded in 96-well plates and were incubated with the indicated concentrations of Dox (**a**) or VP-16 (**b**) plus DMSO or 20 μM of GSK2830371 for 48 hrs. Cell viability was then measured by the CCK-8 assay. Results were represented as % vehicle ± S.D.



**Supplemental Figure S6. GSK2830371 does not have significant effect on mouse body weights.** Mouse body weights at the end of treatment ( $P=0.69$ ) (Student's t-test, two-tailed).