**Supplementary Data**

**Fig. S1. YAP1 overexpression reversed the inhibition in the aggressive behavior of pancreatic cancer cells induced by WDR3 silencing *in vitro***



**A.** Western blot analyses of WDR3 and YAP1 expression in PANC-1 cells infected with sh-Control or sh-WDR3s and/or YAP1. GAPDH served as an internal reference.

**B.** RT-PCR analyses (B) of CTGF and CYR61 expression in PANC-1 cells infected with sh-Control or sh-WDR3s and/or YAP1. GAPDH served as an internal reference. Data are presented as the mean ± SD of three independent experiments. Statistical analyses were performed with one-way ANOVA followed by Tukey's multiple comparison's tests. ns, not significant; \*\*\*, P < 0.001.

**C-E.** PANC-1 infected with sh-Control or sh-WDR3s and/or YAP1 were were harvested for MTS (C), colony formation (D), and Transwell invasion assays (E) after forty-eight hours of culture. Each bar represents the mean ± SD of three independent experiments. Statistical analyses were performed with one-way ANOVA followed by Tukey's multiple comparison's tests. ns, not significant; \*\*\*, P < 0.001.

**Fig. S2. WDR3 expression was positively correlated with YAP1 in pancreatic cancer patient specimens.**



**A.** IHC images of WDR3 and YAP1 staining in TMA tissue sections (n = 31 PDAC samples). The scale bars are shown as indicated.

**B and C.** Heatmap (B) and dot plot (C) showing the correlation of IHC scores for the protein expression of WDR3 and YAP1 in PDAC patient specimens. (r = 0.5916 for Spearman correlation coefficients, P < 0.001).

**D.** Evaluation of the correlation between the expression of WDR3 and YAP1 at the mRNA level in pancreatic cancer samples performed with the GEPIA web tool (P = 1.1e-27, R = 0.7).

**Fig. S3. WDR3 knockdown enhanced the anti-pancreatic cancer effect of YAP1 inhibition.**



**A-C.** Panc02 cells infected with sh-Control or sh-WDR3 #1 were subcutaneously injected into C57BL/6 mice. The mice were treated with TED-347 3 times on days 1, 4 and 7 at a dose of 20 mg/kg. The tumors were harvested and photographed (A) on day 21. Data for tumor volume (B) and tumor mass (C) are shown as the mean ± SD (n = 5). Statistical analyses were performed with two-way ANOVA followed by Sidak's multiple comparison's tests. \*\*\*, P < 0.001.

**Fig. S4. LC-MS/MS assay to identify the base peak of IgG IP and WDR3 IP samples.**



**Fig. S5. GATA4 silencing reversed the induction in the aggressive behavior of pancreatic cancer cells induced by WDR3 overexpression *in vitro***



**A.** RT-PCR analyses (B) of YAP1 expression in PANC-1 cells infected with WDR3s and/or sh-GATA4 #1. GAPDH served as an internal reference. Data are presented as the mean ± SD of three independent experiments. Statistical analyses were performed with one-way ANOVA followed by Tukey's multiple comparison's tests. ns, not significant; \*\*\*, P < 0.001.

**B-D.** PANC-1 infected with WDR3s and/or sh-GATA4 #1 were were harvested for MTS (B), colony formation (C), and Transwell invasion assays (D) after forty-eight hours of culture. Each bar represents the mean ± SD of three independent experiments. Statistical analyses were performed with one-way ANOVA followed by Tukey's multiple comparison's tests. ns, not significant; \*\*\*, P < 0.001.

**Table S1. The primer sequences for RT-qPCR.**

|  |  |  |
| --- | --- | --- |
| Gene | Forward primer (**5**′ - 3′) | Reverse primer (**5**′ - 3′) |
| GAPDH | ATGACAATGAATACGGCTACAGCA | GCAGCGAACTTTATTGATGGTATT |
| WDR3 | ACCAAGCAGTACCTACGCTAT | TTCTCACCACGAAGTGTCACA |
| GATA4 | CGACACCCCAATCTCGATATG | GTTGCACAGATAGTGACCCGT |
| YAP1 | TAGCCCTGCGTAGCCAGTTA | TCATGCTTAGTCCACTGTCTGT |
| CTGF | ACCGACTGGAAGACACGTTTG | CCAGGTCAGCTTCGCAAGG |
| CYR61 | AAACCCGGATTTGTGAGGT | GCTGCATTTCTTGCCCTTT |

**Table S2. The shRNA sequences.**

|  |  |
| --- | --- |
| sh-WDR3 #1 | CCGGAGGACAAGCAGAATCACTATTCTCGAGAATAGTGATTCTGCTTGTCCTTTTTTTG |
| sh-WDR3 #2 | CCGGCCTGGAATACAAGATACTCTTCTCGAGAAGAGTATCTTGTATTCCAGGTTTTTTG |
| sh-GATA4 #1 | CCGGCCAGAGATTCTGCAACACGAACTCGAGTTCGTGTTGCAGAATCTCTGGTTTTTG |
| sh-GATA4 #2 | CCGGGGACATAATCACTGCGTAATCCTCGAGGATTACGCAGTGATTATGTCCTTTTTG |
| sh-Control | Provided by RIBOBIO. |
| si-Control | Provided by RIBOBIO. |
| si-WDR3 #1 and #2 | Provided by RIBOBIO. |

**Table S3. The primer sequences for ChIP-qPCR.**

|  |  |  |
| --- | --- | --- |
| Gene | Forward primer (**5**′ - 3′) | Reverse primer (**5**′ - 3′) |
| **YAP1** | CCGTTTACCCCTCTCAAGTG | CTTAAAGCCGCGAGGATAGA |