

Investigational New Drugs

Structure-Activity Relationship (SAR) of Withanolides to Inhibit Hsp90 for Its Activity in Pancreatic Cancer Cells

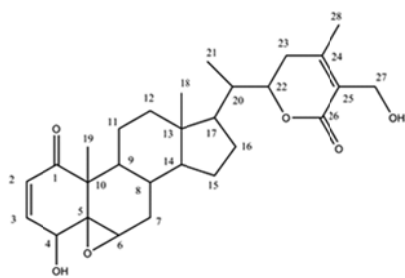
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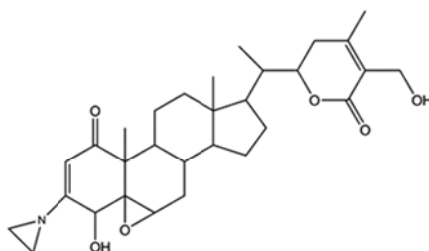
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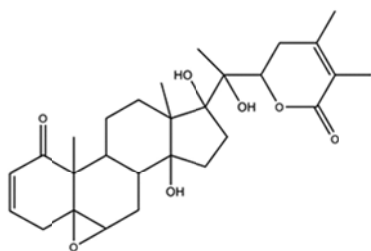
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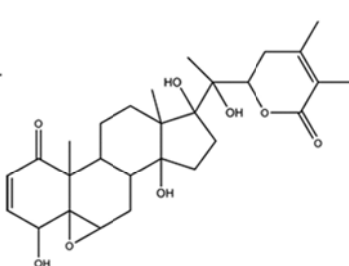
A. Withaferin A (WA)



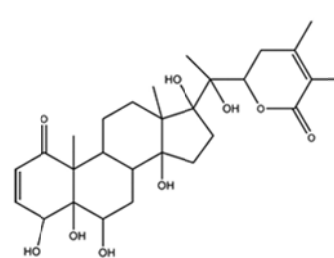
B. 3-Aziridinylwithaferin A (AzWA)



C. Withanolide E (WE)

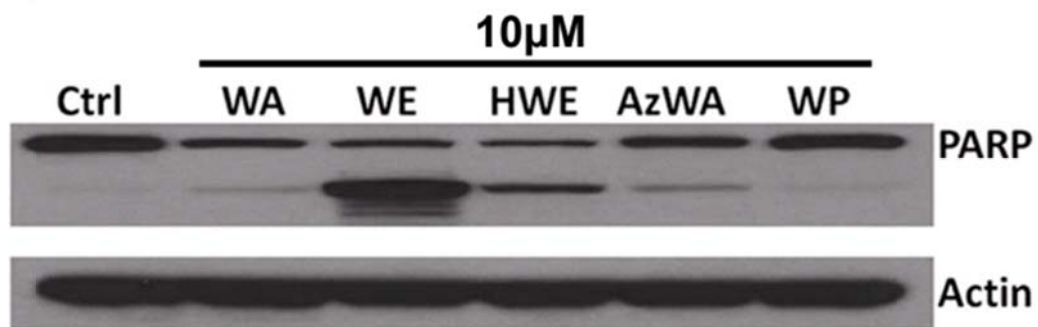


D. 4β-Hydroxywithanolide E (HWE)

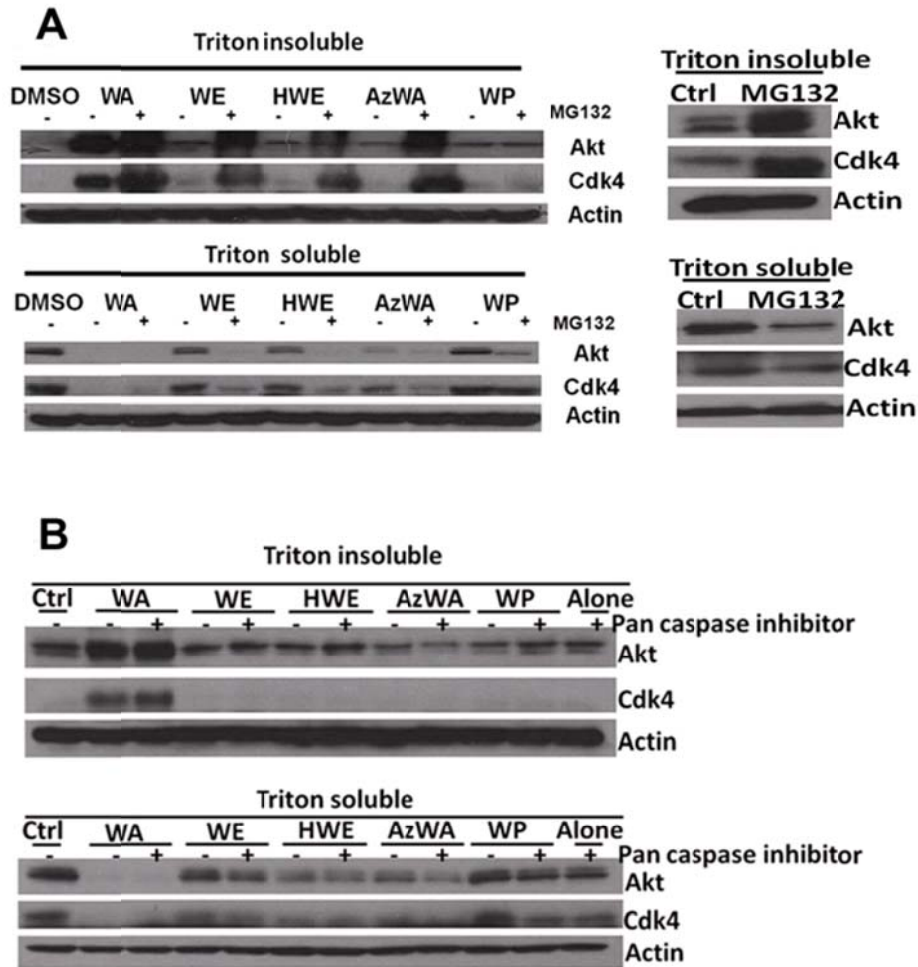


E. Withaperuvin (WP)

S.1 Chemical structure of withaferin A (WA), withanolide E (WE), 4β-hydroxywithanolide E (HWE), 3-Aziridinylwithaferin A (AzWA) and Withaperuvin (WP).



S.2 PARP protein level in Panc-1 cells after 10µM WA, WE, HWE, AzWA or WP treatment for 24h. Equal amounts of protein (50 µg/lane) were subjected to SDS-PAGE and analyzed by Western blot with specific antibodies to PARP and Actin. Actin was served as internal standard. Results are representative of three independent experiments.



S.3 WA and its analogues induced Hsp90 client protein degradation through proteasome-dependent pathway. Panc-1 cells were preincubated with 10 μ M MG132 (A) or Z-VAD-FMK for 1 h, respectively, and then were treated with 5 μ M WA, WE, HWE, AzWA and WP for another 24h. Cells were harvested and lysed in Triton X-100 buffer, and the Triton X-100-insoluble fraction was resolubilized in 2% SDS. Proteins (both triton-soluble and triton-insoluble parts) were subjected to Western blot analysis with specific antibodies to Akt, Cdk4 and Actin. Actin was served as internal standard. Results are representative of three independent experiments.