

## Binding protein analysis of anticancer saponin OSW-1

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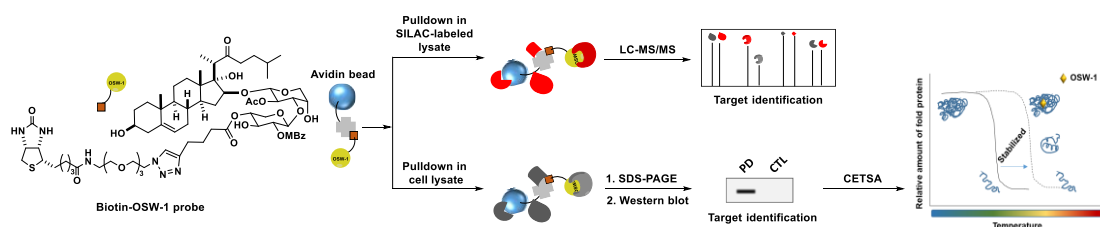
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[Background and aim] Anticancer saponin OSW-1 is a promising chemical tool for uncovering a novel molecular pathway that regulates survival of cancer cells.<sup>1</sup> While its cellular targets are known as OSBP and ORP4,<sup>2</sup> it has been suggested that the compound has other binding proteins. To explore potential cellular targets yet unknown, we previously synthesized biotin-OSW-1 probes for affinity pulldown experiments. In this presentation, we will report the results of the binding protein analysis using chemical probes in combination with the stable isotope labeling by amino acids in cell culture (SILAC)-based approach.<sup>3</sup>

[Methods and results] Biotin-OSW-1 and control probes were each applied to a different set of stable isotope labeled proteins in HeLa cell lysate to obtain quantitative LC-MS/MS analysis data on the pulled-down proteins. These candidate proteins inferred were evaluated by affinity pulldown assay and Western blot analysis. Moreover, we will discuss the data obtained by cellular thermal shift assay (CETSA) as evidence for the binding interaction between OSW-1 and the candidate proteins.



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