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The ARF-AID system: Methods that preserve endogenous protein levels and facilitate rapidly inducible protein degradation

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1 Works for me dx.doi.org/10.17504/protocols.io.bc7ciziw

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ABSTRACT

The ARF-AID (Auxin Response Factor-Auxin Inducible Degron) system is a re-engineered auxin-inducible protein degradation system. Inducible degron systems are widely used to specifically and rapidly deplete proteins of interest in cell lines and organisms. An advantage of inducible degradation is that the biological system under study remains intact and functional until perturbation. This feature necessitates that the endogenous levels of the protein are maintained. However, endogenous tagging of genes with AID can result in chronic, auxin-independent proteasome-mediated degradation. The additional expression of the ARF-PB1 domain in the re-engineered ARF-AID system prevents chronic degradation of AID-tagged proteins while preserving rapid degradation of tagged proteins. Here we describe the protocol for engineering human cell lines to implement the ARF-AID system for specific and inducible protein degradation. These methods are adaptable and can be extended from cell lines to organisms.

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