

Product Data Sheet

Product Name: 5-Ph-IAA

Cat. No.: GC61525

Chemical Properties

Cas. No. 168649-23-8

SMILES O=C(O)CC1=CNC2=C1C=C(C3=CC=CC=C3)C=C2

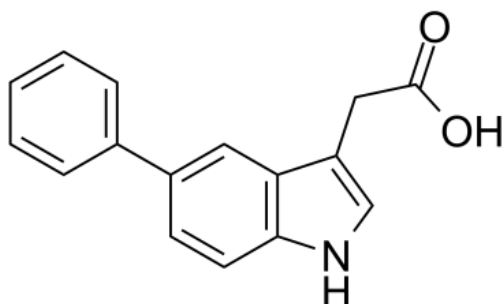
Formula $C_{16}H_{13}NO_2$ M.Wt 251.28

Solubility DMSO: 125 mg/mL (497.45 mM) Storage Store at -20°C

General tips For obtaining a higher solubility, please warm the tube at 37 °C and shake it in the ultrasonic bath for a while. Stock solution can be stored below -20°C for several months.

Shipping Condition Evaluation sample solution: ship with blue ice All other available size: ship with RT, or blue ice upon request.

Structure



Protocol

Cell experiment [1]:

Cell lines	DT40 cells (chicken B lymphoma cell line), HeLa cells (human epithelial immortal cell line), HT1080 cells (human fibrosarcoma cell line), U2OS cells (human osteosarcoma cell line), RPE1 cells (human retinal pigment epithelial cell line), E14tg2a cells (mouse embryonic stem cell line)
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Caution: Product has not been fully validated for medical applications. For research use only.

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Preparation Method	DT40 cells expressing OsTIR1F74A and mScarlet-mAID-CENP-H (or mAID-luciferase, mAID-CENP-A, mAID-CENP-T) were treated with 5-Ph-IAA at indicated concentrations for up to 6h, followed by immunoblotting, luciferase assay, fluorescence microscopy and cell growth/viability assays; HeLa, HT1080, U2OS, RPE1 and E14tg2a cells expressing OsTIR1F74A and mScarlet-mAID-CENP-H were treated with 5 μ M 5-Ph-IAA for 2-4h, followed by immunoblotting, fluorescence microscopy and colony formation assay.
Reaction Conditions	5 μ M; 2-6h
Applications	5-Ph-IAA degraded mScarlet-mAID-CENP-H in OsTIR1F74A-expressing DT40 cells and inhibited cell growth. 5-Ph-IAA degraded mScarlet-mAID-CENP-H in OsTIR1F74A-expressing HeLa, HT1080, U2OS, RPE1 and mouse ES cells within 2-4h, and suppressed colony formation in these cells.
Animal experiment [2]:	
Animal models	Rj:NMRI-Foxn1 nu/nu nude mice (6 weeks old)
Preparation Method	HCT116 (WT ^{AID2} , UHRF1 ^{AID2} , DNMT1 ^{AID2} , UHRF1 ^{mAID2} /DNMT1 ^{mAID2}) cells were suspended in 50% Matrigel and injected subcutaneously into the flanks of mice; when tumors reached ~60mm ³ , mice received daily intraperitoneal injection of 5-Ph-IAA (3mg/kg) or 0.9% NaCl for 8 consecutive days, then tumors were resected for western blot, immunohistochemistry (UHRF1, Ki67, F4/80, p21), whole-mount SA- β -gal staining and tumor weight measurement.
Dosage form	3mg/kg; i.p.; once daily for 8 days

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Applications	5-Ph-IAA degraded UHRF1 and/or DNMT1 in xenograft tumors, reduced tumor weight, decreased Ki67-positive proliferation signal, increased SA-β-gal-positive senescent cells, increased F4/80-positive macrophage infiltration and induced p21 staining in tumor sections.
References: [1] Nishimura K, Yamada R, Hagihara S, et al. A super-sensitive auxin-inducible degron system with an engineered auxin-TIR1 pair. <i>Nucleic Acids Res.</i> 2020 Sep 17;48(18):e108. [2] Chen X, Yamaguchi K, Rodgers B, et al. DNA methylation protects cancer cells against senescence. <i>Nat Commun.</i> 2025 Jul 1;16:5901.	

Background

5-Ph-IAA is a ligand for the AID2 system. 5-Ph-IAA specifically binds to the OsTIR1(F74G) mutant to form a ternary complex. 5-Ph-IAA induces ubiquitination of mini-AID (mAID)-tagged target proteins and degradation by the 26S proteasome. 5-Ph-IAA can be used for dynamic studies of protein function in living cells, single-cell protein isoform studies, and research on colorectal cancer and other tumors^[1-4].

In vitro, treatment of rab1 and rab5 knockout MDCK cells with 1μM 5-Ph-IAA for 2 hours degraded RAB1B and RAB5A proteins in the cells, rendering the cells in RAB1-depleted and RAB5-depleted states, respectively^[5]. Treatment of HCT116 cells (expressing OsTIR1(F74G) and mAID-EGFP-NLS) with 1μM 5-Ph-IAA for 4 hours degraded the mAID-EGFP-NLS reporter protein in the cells^[6]. Treatment of DT40 cells expressing OsTIR1F74A and mScarlet-mAID-CENP-H with 5μM 5-Ph-IAA for 6 hours degraded mScarlet-mAID-CENP-H protein in the cells and inhibited cell growth. Treatment of HeLa, HT1080, U2OS, RPE1 cells and ES cells expressing OsTIR1F74A and mAID-tagged target proteins with 5μM 5-Ph-IAA for 2-4 hours degraded the corresponding mAID-tagged target proteins and inhibited colony formation^[7].

In vivo, a single intraperitoneal injection of 5mg/kg 5-Ph-IAA into adult pMK411 transgenic mice degraded AID-EGFP reporter protein in the heart within 6 hours^[8]. A single intraperitoneal injection

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of 5mg/kg 5-Ph-IAA into Cep192^{AID/AID};OsTir1^{Tir/Tir} mice degraded CEP192-mAID in the small intestine, spleen and stomach. Intraperitoneal injection of 5mg/kg 5-Ph-IAA into Cep192^{wt/wt};OsTir1^{Tir/Tir} mice every 24 hours for 14 consecutive days did not cause behavioral abnormalities, body weight changes, blood and serum toxicity indicator changes or histopathological damage^[9]. Daily intraperitoneal injection of 3mg/kg 5-Ph-IAA into Rj:NMRI-Foxn1 nude mice bearing xenograft tumors formed by HCT116 (WT^{AID2}, UHRF1^{AID2}, DNMT1^{AID2}, UHRF1^{mAID2}/DNMT1^{mAID2}) cells for 8 consecutive days degraded UHRF1 and/or DNMT1 proteins in the tumors, reduced tumor mass, decreased Ki67-positive signal, and increased SA-β-gal-positive signal^[10].

References:

- [1] Sural S, Botero JQ, Hobert O, et al. Protocol to synthesize the auxin analog 5-Ph-IAA for conditional protein depletion in *C. elegans* using the AID2 system. STAR Protoc. 2024 Mar 15;5(1):102901.
- [2] Islam M, Negishi T, Kitamoto N, et al. A single-chain antibody-based AID2 system for conditional degradation of GFP-tagged and untagged proteins. J Cell Sci. 2025 Jul 1;138(13):jcs263961.
- [3] Gameiro E, Juárez-Núñez KA, Fung JJ, et al. Genome-wide conditional degron libraries for functional genomics. J Cell Biol. 2025 Feb 3;224(2):e202409007.
- [4] Shuvalova E, Fortygina P, Smirnova G, et al. PKCβII Activation Promotes Membrane-Proximal Enrichment of Ribosome-Bound RACK1. Int J Mol Sci. 2026 Jun 11;27(12):5310.
- [5] Haga K, Fukuda M. Comprehensive knockout analysis of the RAB family small GTPases reveals an overlapping role of RAB2 and RAB14 in autophagosome maturation. Autophagy. 2025;21(1):21-36.
- [6] Yesbolatova A, Saito Y, Kitamoto N, et al. The auxin-inducible degron 2 technology provides sharp degradation control in yeast, mammalian cells, and mice. Nat Commun. 2020 Nov 11;11(1):5701.
- [7] Nishimura K, Yamada R, Hagihara S, et al. A super-sensitive auxin-inducible degron system with an engineered auxin-TIR1 pair. Nucleic Acids Res. 2020 Sep 17;48(18):e108.

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[8] Makino-Itou H, Yamatani N, Okubo A, et al. Establishment and characterization of mouse lines useful for endogenous protein degradation via an improved auxin-inducible degron system(AID2). Dev Growth Differ. 2024;66(7):384-393.

[9] Sladky VC, Strong MA, Tapias-Gomez D, et al. The AID2 system offers a potent tool for rapid, reversible, or sustained degradation of essential proteins in live mice. bioRxiv. 2024 Jun 5:2024.06.04.597287.

[10] Chen X, Yamaguchi K, Rodgers B, et al. DNA methylation protects cancer cells against senescence. Nat Commun. 2025 Jul 1;16:5901.

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