# $\alpha$ -Amanitin

Cat. No.:	HY-19610
CAS No.:	23109-05-9
Molecular Formula:	C <sub>39</sub> H <sub>54</sub> N <sub>10</sub> O <sub>14</sub> S
Molecular Weight:	918.97
Target:	DNA/RNA Synthesis; ADC Cytotoxin
Pathway:	Cell Cycle/DNA Damage; Antibody-drug Conjugate/ADC Related
Storage:	-20°C, protect from light * The compound is unstable in solutions, freshly prepared is recommended.

# $H_{\text{H}}^{\text{H}}$

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## SOLVENT & SOLUBILITY

Preparing Stock Solutions	1 mM	1.0882 mL	5.4409 mL	10.8817 mL
	5 mM	0.2176 mL	1.0882 mL	2.1763 mL
	10 mM	0.1088 mL	0.5441 mL	1.0882 mL
Please refer to the sol	ubility information to select the ap	propriate solvent.	i	1
1 Add each solvent o	ane hy one: DBS			
	1. Add each solvent o	1. Add each solvent one by one: PBS		

BIOLOGICAL ACTIVITY		
Description	α-Amanitin is the principal toxin of several deadly poisonous mushrooms, exerting its toxic function by inhibiting RNA- polymerase II.	
IC <sub>50</sub> & Target	Traditional Cytotoxic Agents	
In Vitro	$\alpha$ -Amanitin decreases TAF15 mRNA and TAF15 protein levels in MKN45 cells, and inhibits the RNAPII activity towards TAF15 mRNA <sup>[2]</sup> . alpha-Amanitin decreases cell viability by 14%, 21%, 41%, 44%, and 50% at concentrations of 100, 10, 1, 0.1, and 0.01 µg/mL, respectively. The LD <sub>50</sub> of the alpha-Amanitin at 36 h is measured as 1 µg/mL. The total amount of protein within the cell at 24 h is significantly increased for the 1 µg/mL dose of alpha-Amanitin compared to the control <sup>[3]</sup> . $\alpha$ -Amanitin dramatically decreases the expression of gap junctional genes (Gja1, Gja4 and Gjc1) and FSHr and LHr in cumulus cells <sup>[4]</sup> . MCE has not independently confirmed the accuracy of these methods. They are for reference only.	
In Vivo	The intravenous LD <sub>50</sub> dose of alpha-Amanitin is 0.327 mg/kg body weight after intravenous injection into BALB/c mice. After 12 h of alpha-Amanitin injection in caudal vein, the levels of WBC, RBC and HGB decrease significantly, while those of BUN	



and Crea increase significantly in serum. alpha-Amanitin inhibits some genes (Hsp90b1, Irx4, etc.), whose encoded proteins regulate the RNA polymerase II activity. alpha-Amanitin down-regulates some proteins (Nmi, Trpc5, etc.) taking part in the transcription progress<sup>[1]</sup>. alpha-Amanitin has potent activity in DTC suppression. Mice injected with alpha-Amanitin (0.4 mg/kg, i.p.)-treated cells maintain their body weight, while those receiving a peritoneal injection of MKN45 cells show a constant decrease in body weight<sup>[2]</sup>.

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

PROTOCOL	)
Cell Assay <sup>[3]</sup>	The MTT assay is used to evaluate the overall functional integrity and viability of the cultured cells. The MCF-7 cells are put into 96-well plates $(2\times10^4$ for each well), which are incubated for 24 h. The specific concentrations of alpha-Amanitin and $\beta$ - Amanitin are added to the cell culture medium, and plates are incubated for an additional 36 h. MTT solution (1:10 ratio) and dimethyl sulfoxide (DMSO) (100 $\mu$ L) are then added to the cell culture medium and plates are incubated overnight. The absorbance is measured at 570 nm on a plate reader. This experiment is repeated 3 times. The absorbance data are calculated as percentages according to the control group. MCE has not independently confirmed the accuracy of these methods. They are for reference only.
Animal Administration <sup>[2]</sup>	For tumorigenicity tests, six colonies (untreated) and DTCs derived from MKN45 cells are individually injected subcutaneously into the left and right side of the backs of six 6-week-old female nude mice (BALB/cAjcl-nu/nu). These mice are monitored for 49 days after the inoculation or until tumors reach 10 mm in the largest diameter, and are then euthanized. For the PC model, $1.0 \times 10^6$ MKN45 cells are injected intraperitoneally into six 6-week-old female nude mice (BALB/cAjcl-nu/nu). Mice are then treated with CIS (4.0 mg/kg, intraperitoneal administration) or a combination of CIS and alpha-Amanitin (0.4 mg/kg, intraperitoneal administration). For the combination treatment, alpha-Amanitin is given 24 hours before CIS. Body weight is monitored for 28 days after the treatment. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

### CUSTOMER VALIDATION

- Mol Cancer. 2023 Apr 27;22(1):77.
- Mol Cancer. 2019 Nov 21;18(1):167.
- J Hematol Oncol. 2021 Mar 19;14(1):46.
- Nat Commun. 2023 Feb 16;14(1):863.
- Nat Commun. 2018 Apr 30;9(1):1726.

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### REFERENCES

[1]. Zhao J, et al. Pathological effects of the mushroom toxin alpha-amanitin on BALB/c mice. Peptides. 2006 Dec;27(12):3047-3052.

[2]. Kume K, et al. α-Amanitin Restrains Cancer Relapse from Drug-Tolerant Cell Subpopulations via TAF15. Sci Rep. 2016 May 16;6:25895

[3]. Kaya E, et al. Evaluation and comparison of alpha- and beta-amanitin toxicity on MCF-7 cell line. Turk J Med Sci. 2014;44(5):728-32

[4]. Park MW, et al. RNA Polymerase II Inhibitor, α-Amanitin, Affects Gene Expression for Gap Junctions and Metabolic Capabilities of Cumulus Cells, but Not Oocyte, during in vitro Mouse Oocyte Maturation. Dev Reprod. 2013 Mar;17(1):63-72

### Caution: Product has not been fully validated for medical applications. For research use only.

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