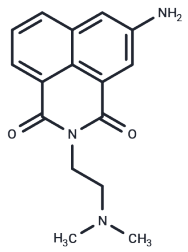
Data Sheet (Cat.No.T6336)

TargetMol

Amonafide

Chemical Properties



CAS No. : 69408-81-7

Formula: C16H17N3O2

Molecular Weight: 283.33

Storage: Powder: -20°C for 3 years | In solvent: -80°C for 1 year
Actual storage temperature shall be subject to the COA.

Biological Description

Description	Amonafide (NSC-308847,AS1413)(AS1413) produces protein-associated DNA-strand breaks through a topoisomerase II-mediated reaction, but does not produce topoisomerase I-mediated DNA cleavage.
Targets(IC50)	Topoisomerase
In vitro	Through a topoisomerase II-mediated reaction, Amonafide treatment produces DNA single-strand breaks (SSB), double-strand breaks (DSB), and DNA-protein cross-links in human myeloid leukemia cells. Amonafide treatment inhibits conlony formation of the leukemic cell lines and the normal human bone marrow GM-CFC in a dose-dependent manner. Amonafide does not produce topoisomerase I-mediated DNA cleavage even at 100 μM. The m-AMSA-resistant line is less than 2-fold resistant to Amonafide [1] Amonafide interferes with the DNA breakage-reunion activity of mammalian DNA topoisomerase II resulting in DNA cleavage stimulation. [2] Compared with those of other antitumor drugs, Amonafide-stimulated cleavage intensity patterns are markedly different. Amonafide highly prefers a cytosine, and excludes guanines and thymines instead, at position -1, with lower preference for an adenine at position +1. [3] Topoisomerase II-mediated DNA cleavage induced by Amonafide is affected only slightly (less than 3-fold) by 1 mM ATP, suggeting that Amonafide is an ATP-insensitive topoisomerase II inhibitor in contrast to doxorubicin, etoposide, and mitoxantrone. [4] Amonafide significantly inhibits the growth of HT-29, HeLa, and PC3 cells with IC50 of 4.67 μM, 2.73 μM, and 6.38 μM, respectively. [5] Amonafide is unaffected by P-glycoprotein-mediated efflux, unlike those of the classical topoisomerase II inhibitors (daunorubicin, doxorubicin, idarubicin, etoposide, and mitoxantrone). [6]
Cell Research	All cell lines are in the logarithmic phase of growth when the assay of 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide (MTT) is carried out. Cells are harvested and seeded into 96-well tissue culture plates at a density of 2.5 × 103 cells/well in 150 μL aliquots of medium. The concentrations tested are serial dilutions of a stock solution (10 μM in DMSO) with phosphate-buffered saline (PBS) and are added 24 hours later. The assay is ended after 72 hours of Amonafide exposure and PBS is used as a negative control. After 72 hours treatment, cells are washed twice with PBS, and then 50 μL/well of MTT reagent (1 mg/mL in PBS) together with 150 μL/well of prewarmed medium are added. The plates are returned to the incubator for 4 hours. Subsequently, DMSO is added as solvent. Absorbance is determined at 570 nm with a Microplate reader. All experiments are performed at least three times, and the average

Cell Research	of the percentage absorbance is plotted against concentration. Then, the concentration of Amonafide required to inhibit 50% of cell growth (IC50) is calculated for Amonafide. (Only for Reference)

Solubility Information

Solubility	DMSO: 53 mg/mL (187.06 mM),Sonication is recommended. Ethanol: 4 mg/mL (14.12 mM),Sonication is recommended. H2O: < 1 mg/mL (insoluble or slightly soluble), (< 1 mg/ml refers to the product slightly soluble or insoluble)
In vivo Formulation	10% DMSO+40% PEG300+5% Tween 80+45% Saline: 2 mg/mL (7.06 mM),Sonication is recommended. *Please add the solvents sequentially, clarifying the solution as much as possible before adding the next one. Dissolve by heating and/or sonication if necessary. Working solution is recommended to be prepared and used immediately. The formulation provided above is for reference purposes only. In vivo formulations may vary and should be modified based on specific experimental conditions.*

Preparing Stock Solutions

	1mg	5mg	10mg
1 mM	3.5295 mL	17.6473 mL	35.2945 mL
5 mM	0.7059 mL	3.5295 mL	7.0589 mL
10 mM	0.3529 mL	1.7647 mL	3.5295 mL
50 mM	0.0706 mL	0.3529 mL	0.7059 mL

Please select the appropriate solvent to prepare the stock solution, according to the solubility of the product in different solvents. Please use it as soon as possible.

Note: The dilution table applies only to solid products. For liquid products, please calculate the stock solution based on the stated concentration and/or density.

Reference

Andersson BS, et al. Cancer Res, 1987, 47(4), 1040-1044.
Hsiang YH, et al. Mol Pharmacol, 1989, 36(3), 371-376.
De Isabella P, Nucleic Acids Res, 1995, 23(2), 223-229.
Wang H, et al. J Biol Chem, 2001, 276(19), 151990-151995.
BrañaMF, et al. J Med Chem, 2004, 47(6), 1391-1399.