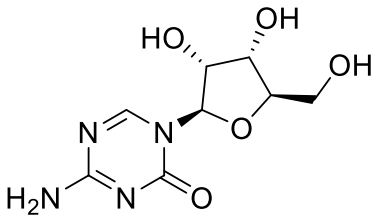
Product data sheet

MedKoo Cat#: 100070 Name: Azacitidine CAS#: 320-67-2 Chemical Formula: $C_8H_{12}N_4O_5$ Exact Mass: 244.08077 Molecular Weight: 244.2	
Product supplied as:	Powder
Purity (by HPLC):	≥ 98%
Shipping conditions	Ambient temperature
Storage conditions:	Powder: -20°C 3 years; 4°C 2 years.
	In solvent: -80°C 3 months; -20°C 2 weeks.

1. Product description:
Azacitidine is a pyrimidine nucleoside analogue of cytidine with antineoplastic activity. Azacitidine is incorporated into DNA, where it reversibly inhibits DNA methyltransferase, thereby blocking DNA methylation. Hypomethylation of DNA by azacitidine may activate tumor suppressor genes silenced by hypermethylation, resulting in an antitumor effect. This agent is also incorporated into RNA, thereby disrupting normal RNA function and impairing tRNA cytosine-5-methyltransferase activity.

2. CoA, QC data, SDS, and handling instruction
SDS and handling instruction, CoA with copies of QC data (NMR, HPLC and MS analytical spectra) can be downloaded from the product web page under "QC And Documents" section. Note: copies of analytical spectra may not be available if the product is being supplied by MedKoo partners. Whether the product was made by MedKoo or provided by its partners, the quality is 100% guaranteed.

3. Solubility data

Solvent	Max Conc. mg/mL	Max Conc. mM
DMSO	31	126.95
Water	33.33	136.49

4. Stock solution preparation table:

Concentration / Solvent Volume / Mass	1 mg	5 mg	10 mg
1 mM	4.10 mL	20.48 mL	40.95 mL
5 mM	0.82 mL	4.10 mL	8.19 mL
10 mM	0.41 mL	2.05 mL	4.10 mL
50 mM	0.08 mL	0.41 mL	0.82 mL

5. Molarity Calculator, Reconstitution Calculator, Dilution Calculator
Please refer the product web page under section of "Calculator"

6. Recommended literature which reported protocols for in vitro and in vivo study
In vitro study

1. Polansky JK, Kretschmer K, Freyer J, Floess S, Garbe A, Baron U, Olek S, Hamann A, von Boehmer H, Huehn J. DNA methylation controls Foxp3 gene expression. Eur J Immunol. 2008 Jun;38(6):1654-63. doi: 10.1002/eji.200838105. PMID: 18493985.

2. Osorio-Montalvo P, Sáenz-Carbonell L, De-la-Peña C. 5-Azacytidine: A Promoter of Epigenetic Changes in the Quest to Improve Plant Somatic Embryogenesis. Int J Mol Sci. 2018 Oct 16;19(10):3182. doi: 10.3390/ijms19103182. PMID: 30332727; PMCID: PMC6214027.

In vivo study

1. Travers M, Brown SM, Dunworth M, Holbert CE, Wiehagen KR, Bachman KE, Foley JR, Stone ML, Baylin SB, Casero RA Jr, Zahnow CA. DFMO and 5-Azacytidine Increase M1 Macrophages in the Tumor Microenvironment of Murine Ovarian Cancer. Cancer Res. 2019 Jul 1;79(13):3445-3454. doi: 10.1158/0008-5472.CAN-18-4018. Epub 2019 May 14. PMID: 31088836; PMCID: PMC6606334.

7. Bioactivity

Product data sheet

Biological target:

Azacitidine (5-Azacytidine, 5-AzaC, Ladakamycin, AZA, 5-Aza, CC-486) is a nucleoside analogue of cytidine that specifically inhibits DNA methylation by trapping DNA methyltransferases.

In vitro activity

To test whether efficient DNA demethylation is indeed critical for the induction of stable Foxp3 expression in vitro, DNA-hypomethylating Aza (azacitidine, 5-azacytidine or 5-aza-deoxycytidine) was used to interfere with de novo DNA methylation. During replication these nucleoside analogs are integrated into DNA (and RNA for 5-azacytidine) and subsequently interfere with the function of DNMT1 (DNA methyltransferase 1) leading to rapid passive DNA demethylation. To test the impact of DNA methylation on the stability of Foxp3 expression, Treg was first indued in vitro by activation of Foxp3–CD25–CD4+ T cells in the presence of TGF-β. After sorting of Foxp3+ iTreg to high purity (97%), cells were restimulated either alone or in the presence of TGF-β or Aza. As depicted in Fig. 2, Aza significantly increased the proportion of cells expressing Foxp3 compared to restimulation cultures without exogenous TGF-β and Aza, which rapidly lost Foxp3 expression over time (Fig. 2B). On day 4 of restimulation, the percentage of Foxp3+ cells in Aza-treated cultures was more than 4-fold increased (Fig. 2A, 47% and 9.3% cells maintained Foxp3 expression in Aza-treated and non-treated cultures, respectively). TGF-β-containing control cultures maintained high Foxp3 expression with 91% Foxp3+ cells on day 4 (Fig. 2A). These results strengthen the idea of an essential role for DNA (de)methylation in the regulation of Foxp3 stability.

Reference: Eur J Immunol. 2008 Jun;38(6):1654-63. https://doi.org/10.1002/eji.200838105

In vivo activity

Immunocompetent C57BL/6 mice were injected intraperitoneally (i.p.) with 250,000 VDID8 syngeneic MOSE cells. Mice were treated IP with azacitidine (AZA) (0.5 mg/kg). Hemorrhagic ascites fluid consistently develops at approximately 4–5 weeks post VDID8 injection and is an accurate measurement of tumor burden in mice, allowing observation of tumor growth in real time. After draining hemorrhagic ascites fluid from mice for the second time (typically week 5 post tumor injection), mice treated with single agent AZA demonstrated significantly lower tumor burdens (Fig 1b). Mice treated with AZA had an increase in median survival of 44 days (Fig 1c). Total numbers of lymphocytes are significantly increased by single agent AZA (Fig 1d,,ee). AZA led to significant increases in T cell, NK cell, and IFNγ+ lymphocyte populations examined in the tumor microenvironment (Fig 2a–g).

Reference: Cancer Res. 2019 Jul 1;79(13):3445-3454. https://www.ncbi.nlm.nih.gov/pmc/articles/pmid/31088836/