

# **Bleomycin Sulfate**

Prin

#### **Technical Data**

Molecular Weight	1512.62	Storage	3 years -20°C powder
Formula	C <sub>55</sub> H <sub>85</sub> N <sub>17</sub> O <sub>25</sub> S <sub>4</sub>		2 years -80°C in solvent
CAS No.	9041-93-4	Synonyms	Blenoxane
Solubility (25°C) *	In vitro	DMSO	100 mg/mL warmed (66.11 mM)
		Water	100 mg/mL (66.11 mM)
		Ethanol	Insoluble
	In vivo	Saline	30 mg/mL

<sup>\*&</sup>lt;1 mg/ml means slightly soluble or insoluble.

\* Please note that Selleck tests the solubility of all compounds in-house, and the actual solubility may differ slightly from published values. This is normal and is due to slight batch-to-batch variations.

Chemical Name no
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#### **Preparing Stock Solutions**

Volume (DMSO) Mass Concentration	1 mg	5 mg	10 mg
1 mM	0.6611 mL	3.3055 mL	6.6110 mL
5 mM	0.1322 mL	0.6611 mL	1.3222 mL
10 mM	0.0661 mL	0.3306 mL	0.6611 mL
50 mM	0.0132 mL	0.0661 mL	0.1322 mL

## **Biological Activity**

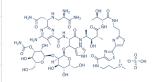
Description	Bleomycin Sulfate is a glycopeptide antibiotic and an anticancer agent for squamous cell carcinomas (SCC) with IC50 of 4 nM in UT-SCC-19A cells.
In vitro	UT-SCC-12A and UT-SCC-12B are both more resistant to Bleomycin sulfate with IC50 of 14.2 nM and 13 nM, respectively.  Alveolar macrophages incubated with 0.01 μg/mL to 1μg /mL Bleomycin sulfate for 18 hours secretes significantly more fibroblast growth factor than macrophages incubated without Bleomycin sulfate. Macrophages stimulated with Bleomycin sulfate continues to produce significant amounts of fibroblast growth factor even after Bleomycin sulfate is removed and replaced with fresh (Bleomycin sulfate-free) media. Fibroblast growth factor secretion by Bleomycin sulfate-stimulated alveolar macrophages is inhibited by cycloheximide, and the 5-lipoxygenase inhibitors NDGA (nordihydroguaiaretic acid) and BW755c, indicating not only a requirement for protein synthesis but also for metabolites of the 5-lipoxygenase pathway of arachidonic acid metabolism for full expression of activity. <sup>[2]</sup> Bleomycin sulfate (400 μg/mL) incubation for 24 hours decreases the viability of NTera-2 cells, and increases caspase-3, -8 and -9 activities, Bax and cytoplasmic cytochrome c levels and decreases Bcl-2 levels. <sup>[3]</sup> In terms of unstable aberrations, the clastogenic effect of Bleomycin sulfate on ADIPO-P2 cells persists for at least 10 days after exposure. Bleomycin sulfate-induced telomere instability in mammalian cells persists for several generations after exposure. Moreover, the appearance of telomere fusions in Bleomycin sulfate-exposed cells 10 days after treatment suggests that Bleomycin sulfate can induce delayed telomere instability. <sup>[4]</sup>
In vivo	Day 7 post-Bleomycin sulfate, CD45+ cells in BALf in NOX-/- is 1.7-fold > WT, 57% of which are Mf that decreases by 67% in WT and 83% in NOX-/- by Day 21. [5]
Features	

### Protocol (Only for Reference)

# Cell Assay:

Cell lines	ADIPO-P2 cells
Concentrations	2.5 μg/mL
Incubation Time	30 minutes
Method	ADIPO-P2 cells are grown in D-MEM high glucose medium supplemented with 20% fetal calf serum, penicillin (100 U/mL) and streptomycin (100 µg/mL) at 37 °C and 5% CO <sub>2</sub> atmosphere. Cells are cultured as monolayer in TC25 Corning flasks containing 1.5 × 10 <sup>5</sup> cells/mL. For each experiment, two flasks are set up, one for the control and one for the treated culture. During the log phase of growth ADIPO-P2 cells are treated with a 30 minutes pulse of 2.5 µg/mL of Bleomycin sulfate. Control cultures are set up in parallel but not exposed to Bleomycin sulfate. Time of exposure and concentration of Bleomycin sulfate are chosen according to previous studies carried out in our laboratory with mammalian cells exposed to Bleomycin sulfate. At the end of the pulse treatment with Bleomycin sulfate, the cells are washed twice with Hank's balanced salt solution and kept in culture with fresh culture medium until harvesting. Cells are continuously maintained in culture during 5 passages or

### **Chemical Structure**



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subcultures after treatment. Subcultivation is carried out whenever the cultures became confluent (approximately  $4 \times 10^5$ cells/mL of culture medium). To estimate cell growth, at the time of subcultivation cells are collected by trypsinization, an aliquot of about 200  $\mu$ L stained with 0.4% trypan blue, and the number of viable cells is determined. Cells are then suspended in fresh culture medium and dispensed into new culture flasks containing 1 × 10<sup>5</sup> cells/mL to continue growing. The rest of the cells is discarded or dispensed in another flask for cytogenetic analysis, which is performed at 18 hours and 10 days after the end of treatments. To analyze chromosomal aberrations, colchicine (0.1 µg/mL) is added to cell cultures during the last 3 hours of culture. Chromosome preparations are made following standard procedures. After harvesting, cells are hypotonically shocked, fixed in methanol:acetic acid (3:1), spread onto glass slides and processed for PNA-FISH. Two independent experiments are carried out.

# Animal Study:

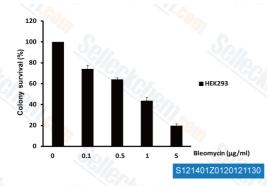
Animal Models	CD-1 mice
Formulation	Saline
Dosages	5 mg/kg, 2 ml/kg
Administration	Administered via i.t.

#### References

- [1] Jääskelä-Saari HA, et al. Acta Otolaryngol Suppl. 1997, 529, 241-244. [2] Denholm EM, et al. Am J Pathol. 1989, 134(2), 355-363.
- [3] Cort A, et al. Mol Med Report. 2012, 5(6), 1481-1486. [4] Paviolo NS, et al. Mutat Res. 2012, 734(1-2), 5-11. [5] Banerjee ER, et al. Stem Cell Res Ther. 2012, 3(3), 21.

- [6] Danesi CC, et al. Mutat Res. 2012.
- [7] Shi K, et al. Int J Clin Exp Med. 2014, 7(9), 2645-2650.

#### **Customer Product Validation**



Data from [Data independently produced by AACRElizabeth williamson from university of florida., 2011]

## Bleomycin Sulfate has been referenced in 6 publications.

Increased mutagen sensitivity and DNA damage in Pulmonary Arterial Hypertension [Federici C, et al. Am J Respir Crit Care Med, 2015, 10.1164/rccm.201411-2128OC]	PubMed: 25918951
The nuclease FAN1 is involved in DNA crosslink repair in Arabidopsis thaliana independently of the nuclease MUS81. [Herrmann NJ, et al. Nucleic Acids Res, 2015, 10.1093/nar/gkv208]	PubMed: 25779053
MHF1 plays FANCM-dependent and -independent roles in DNA repair and homologous recombination in plants. [Dangel NJ, et al. Plant J, 2014, 78(5):822-33]	PubMed: 24635147
Nuclear PTEN interferes with binding of Ku70 at double-strand breaks through post-translational poly (ADP-ribosyl) ation. [Guan J, et al. Biochim Biophys Acta, 2016, 1863(12):3106-3115]	PubMed: 27741411
MCM8-9 complex promotes RAD51 recruitment at DNA damage sites to facilitate homologous recombination. [Park J, et al. Mol Cell Biol, 2013, 33(8):1632-44]	PubMed: 23401855
Gene-drug interactions and the evolution of antibiotic resistance [Adam Christopher Palmer , et al. Harvard University, 2012, Massachusetts]	

## PLEASE KEEP THE PRODUCT UNDER -20°C FOR LONG-TERM STORAGE.

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