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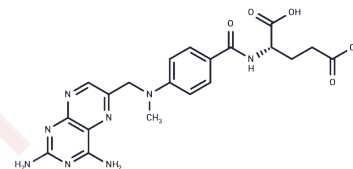
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## Methotrexate

## Chemical Properties

CAS No. :	59-05-2
Formula:	C <sub>20</sub> H <sub>22</sub> N <sub>8</sub> O <sub>5</sub>
Molecular Weight:	454.44
Appearance:	no data available
Storage:	keep away from direct sunlight Powder: -20°C for 3 years   In solvent: -80°C for 1 year



## Biological Description

Description	Methotrexate (WR19039) is a folate analog, an inhibitor of the dihydrofolate reductase DHFR. Methotrexate has antimetabolic, antitumor, and immunosuppressive activities, and is commonly used in rheumatoid arthritis and various tumors.
Targets(IC50)	Apoptosis,Dehydrogenase,DNA/RNA Synthesis,Antifolate,ADC Cytotoxin
In vitro	<p><b>METHODS:</b> Six pediatric leukemia and lymphoma cell lines, NALM-6, NALM-16, JURKAT, CEM, RAMOS, and NAMALWA, were treated with Methotrexate (0.002-5 <math>\mu</math>M) for 120 h, and the proliferation inhibitory activity of the cells was assayed using the SRB method.</p> <p><b>RESULTS:</b> The median IC<sub>50</sub> of Methotrexate against tumor cells was 78 nM, and the IC<sub>50</sub> of the six tumor cells ranged from 33-133 nM. [1]</p> <p><b>METHODS:</b> Human ovarian cancer cells SKOV-3 were treated with Methotrexate (15-50 <math>\mu</math>M) for 24 h. Apoptosis was detected using AO/EtBr probe.</p> <p><b>RESULTS:</b> Methotrexate induced apoptosis in SKOV-3 cells. [2]</p>
In vivo	<p><b>METHODS:</b> To test for chronic toxicity, Methotrexate (0.25-6 mg/kg) was administered intraperitoneally to C57BL/6, DBA/2, and C3H mice five times per week for 12-18 months.</p> <p><b>RESULTS:</b> The 0.25-2 mg/kg dose was well tolerated with minimal cytostasis in lymphoid tissues, testis and skin. 3-6 mg/kg dose produced well-known acute to subacute hematopoietic and gastrointestinal injuries leading to early death. [3]</p> <p><b>METHODS:</b> To examine the mode of action in different types of rheumatoid arthritis (RA) and multiple sclerosis (MS) models, Methotrexate (0.1-5 mg/kg) was injected intraperitoneally once a day for fourteen days into RA and MS models with different pathogenesis.</p> <p><b>RESULTS:</b> Methotrexate showed strong ameliorative effects in classical RA models such as CIA and PIA as well as in MS and EAE models. Methotrexate acts only prior to and dependent on T-cell activation in T-cell activated diseases. [4]</p>
Cell Research	Methotrexate (MTX) is dissolved in DMSO and stored, and then diluted with appropriate media before use[1]. Each cell line is studied in growth inhibition experiments using 96-well microtiter plates. As antifols are schedule dependent, preliminary experiments are aimed at defining the longest duration of exposure that would allow for continuous logarithmic phase growth of cells without changing of the culture media while maintaining a linear relationship between SRB optical density and cell number. Twenty-four hours after cell plating, the cell lines are exposed to the antifol for 120 h (three replicates per experiment). To ensure that a complete sigmoidal survival-concentration

curve could be observed, the following drug concentrations are studied: Methotrexate (0.002-5  $\mu$ M), AMT (0.0001-1  $\mu$ M), PXD (0.0003-10  $\mu$ M), TLX (0.0002-0.5  $\mu$ M). Experiments are repeated at least twice[1].

### Solubility Information

Solubility DMSO: 45 mg/mL (99.02 mM),  
Ethanol: < 1 mg/mL (insoluble or slightly soluble),  
< 1 mg/ml refers to the product slightly soluble or insoluble)

### Preparing Stock Solutions

	1mg	5mg	10mg
1 mM	2.2005 mL	11.0026 mL	22.0051 mL
5 mM	0.4401 mL	2.2005 mL	4.401 mL
10 mM	0.2201 mL	1.1003 mL	2.2005 mL
50 mM	0.044 mL	0.2201 mL	0.4401 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.

### Reference

Norris RE, et al. Clinical potency of methotrexate, aminopterin, talotrexin and pemetrexed in childhood leukemias. Cancer Chemother Pharmacol. 2010 May;65(6):1125-30.

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