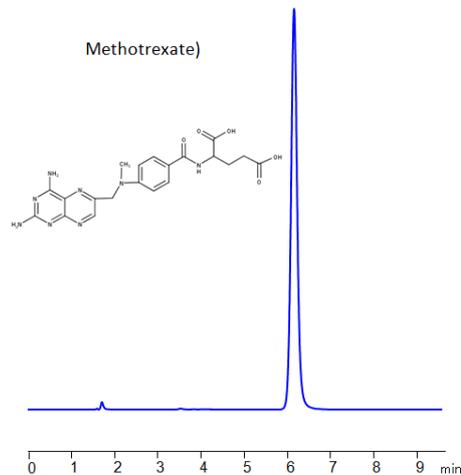


## HPLC Method for Analysis of Methotrexate on Primesep 100 Column



<b>Column:</b>	Primesep 100
<b>Column size:</b>	4,6 × 150 mm, 5 µm
<b>Column part number:</b>	100-46.150.0510
<b>Mobile phase:</b>	MeCN/H <sub>2</sub> O – 50/50%
<b>Buffer:</b>	Ammonium formate pH 3.0 – 20 mM
<b>Flow rate:</b>	1.0 mL/min
<b>Detection:</b>	UV 300 nm

Separation type: Liquid Chromatography Mixed-mode

Methotrexate is a chemotherapy agent and immune system suppressant. It is classified as an antimetabolite and antifolate drug. Methotrexate works by inhibiting the enzyme dihydrofolate reductase (DHFR), preventing the formation of active folate and thus interfering with DNA and RNA synthesis.

Due to its mechanism of action, methotrexate is used to treat certain types of cancer, such as leukemia, breast cancer, lung cancer, and osteosarcoma. It is also used in the treatment of autoimmune diseases, including rheumatoid arthritis and psoriasis, as it can decrease the activity of the immune system.

Like all medications, methotrexate can cause side effects and should be used under the supervision of a healthcare provider. Always consult with a healthcare professional for the most accurate and up-to-date information.

This is a chemotherapy agent and immune system suppressant. Methotrexate is an antimetabolite and antifolate drug, meaning it inhibits the metabolism of folic acid. It does this by inhibiting the enzyme dihydrofolate reductase (DHFR), preventing the formation of active folate and thus interfering with DNA and RNA synthesis. It is used to treat certain types of cancer and autoimmune diseases like rheumatoid arthritis and psoriasis.

Methotrexate retained and analyzed using a reverse-phase Primesep 100, 4.6 x 150 mm, 5 µm, 100 Å, dual ended column. The mobile phase for this method consists of water, acetonitrile (MeCN), and ammonium formate, which serves as a buffer. This analytical method can be monitored using UV detection at 290 nm, an Evaporative Light Scattering Detector (ELSD), or any other evaporative detection method such as Charged Aerosol Detection (CAD) or Electrospray Ionization Mass Spectrometry (ESI-MS)

LOD was determined for this combination of instrument, method, and analyte, and it can vary from one laboratory to another even when the same general type of analysis is being performed.

### Method Parameters

<b>Column</b>	Primesep 100, 4.6 x 150 mm, 5 µm, 100 Å, dual ended
<b>Mobile Phase</b>	MeCN/H <sub>2</sub> O -50/50%
<b>Buffer</b>	Ammonium formate pH 3.0 – 20 mM
<b>Flow Rate</b>	1.0 mL/min
<b>Detection</b>	UV 290 nm
<b>Limit of Detection</b>	34 ppb
<b>Injection Volume</b>	5 µl

Quelle: <https://sielc.com/hplc-separation-of-methotrexate>