Pharmacokinetics of High-Dose Methotrexate

Kristine Radomski Crews, Pharm.D.
St. Jude Children’s Research Hospital
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Overview

- Pharmacokinetics of high-dose methotrexate
- Therapeutic drug monitoring to avoid high-dose methotrexate toxicity
Definition of High-Dose Methotrexate (HDMTX)

- MTX Dose > 500 mg/m²
- Requires leucovorin rescue to prevent severe toxicity
Review of Methotrexate Pharmacology

- MTX inhibits dihydrofolate reductase and blocks de novo nucleotide synthesis by depleting cells of reduced tetrahydrofolate cofactors
- MTX and dihydrofolates can inhibit thymidylate synthase and other enzymes involved in the purine pathway
- Leucovorin restores reduced folate pool
Methotrexate Distribution

- Distributes into kidneys, liver, gastrointestinal tract, muscle, erythrocytes
- *Most important aspect: ability to concentrate into ascites, pleural effusions*
  - “Third-spacing” - causes a sustained release effect as drug releases into general circulation
Methotrexate undergoes active transport

- Active transport into cells
  - This process is saturable and subject to competition with leucovorin
  - Higher leucovorin doses are required when MTX concentrations are high to overcome selective active transport into cell
Major Metabolites of MTX

- DAMPA- formed in GI tract by bacteria, excreted into feces
- 7-OH MTX- little activity; competes w/ MTX for protein binding sites, active transport
- Polyglutamate metabolites- formed intracellularly and accumulate within cell
  - Active metabolites
Methotrexate Excretion

- 60-90% renal
- Active secretion component - saturable process
- Primary route: filtration
- Methotrexate clearance is correlated with GFR/creatinine clearance
Dose-Limiting Toxicity of HDMTX

- Myelosuppression
- Gastrointestinal mucositis
- Nausea/vomiting
- Renal dysfunction – keep pt well-hydrated and increase urine pH with sodium bicarbonate to avoid MTX precipitating out in kidney
Pharmacokinetic Metrics
Assessing Systemic Exposure

AUC highly correlated with concentration at end of infusion
Goals for Therapeutic Drug Monitoring of HDMTX

- Toxicity monitoring related to MTX Cp
  - Leucovorin rescue

- Antitumor efficacy related to MTX exposure (either Cp or AUC)
  - Controversial
Therapeutic Drug Monitoring to Predict and Prevent High-Dose MTX Toxicity

- Survey of cancer treatment centers in US
  - Severe myelosuppression and mucositis developed in ~10% of patients
  - 6% fatality rate attributable to drug toxicity (29/498 patients)

- Stoller et al postulated that routine monitoring of MTX levels would predict early detection of patients at high risk of toxicity

Early identification (24-36 hrs post infusion) and determination of elimination rate permits initiation of a modified leucovorin rescue to prevent toxicity

- MTX effects may not be adequately rescued if delayed 42-48 hrs
- Cytotoxicity function of concentration and duration of exposure

Evans, *Cancer Chemother Pharmacol*, 1979
Patient Characteristics Associated with Toxicity following High-Dose Methotrexate

- Routine monitoring of MTX levels predicts early detection of patients at high risk of toxicity:
  - High 24-hr [MTX]
  - Low urine pH recorded at anytime during the MTX infusion
  - Occurrence of emesis during the infusion

Stoller, *NEJM*, 1977
General Concepts of HDMTX-Leucovorin Rescue

- **Goal of monitoring for toxicity**
  - Ensure that all pts receive adequate leucovorin to prevent severe toxicity (but not “over-rescue”)
- **Early studies showed sustained [MTX] associated with toxicity (after 12 g/m$^2$):**
  - 24 hr > 10 μM
  - 48 hr > 1 μM
  - 72 hr > 0.1 μM
- **In absence of elevated [MTX], follow “standard” leucovorin rescue and toxicity minimal**
Plasma Clearance of High-Dose Methotrexate

Goal: >1000 µM
Goal: <10 µM
Goal: <1 µM

Fig 8. Plasma disappearance curve of 1,045 HD-MTX therapies (12 g/m² MTX/4 hours). At time 0, 4, 12, 24, 36, 48, and 72 hours after the end of the MTX infusion, the mean value ± 1 SD of the MTX concentration is shown.
Guidelines for Administering High-Dose Methotrexate

- Avoid administering MTX with the following drugs:
  - Nephrotoxins:
    - Amphotericin
    - Acyclovir
    - Non-steroidal anti-inflammatory drugs
  - Drugs that can compete with MTX excretion:
    - Penicillins
    - Omeprazole
    - Azole antifungals
Hydration: pre-hydrate with sodium bicarbonate beginning the evening prior to administration of HDMTX

- **D$_5$W$\frac{1}{4}$ NS + Na Bicarbonate 40 mEq/L + KCl 10 mEq/L
- IV fluids should run at a rate of $\geq$100 mL/m$^2$/hr for a minimum of 12 hours prior to starting HDMTX.

Check urine pH with each void

- A urinary pH $\geq$ 6.5 must be achieved before starting MTX infusion

Maintain until serum MTX level is $< 1 \mu$M
**Methotrexate-Leucovorin Guidelines for St Jude OS2008**

**MTX Levels**

Obtain MTX levels at the end of infusion (Hr 4), 24 and 48 hrs from start of the infusion.

- 4 hr MTX level goal: 1000 microM; if >1500 microM, keep IVF rate at 200 mL/m²/hr until 24 hr level known.
- 24 hr MTX level goal: < 10 microM; if > 10 microM, adjust IVF hydration rate and leucovorin per protocol.
- Ensure strict I/O, repeat serum chemistries.
- 48 hr MTX level goal: < 1 microM; if > 1 microM, adjust IVF hydration rate and leucovorin per protocol.
- 72 hr MTX level goal: < 0.1 microM; if > 0.1 microM, adjust IVF fluid hydration and leucovorin per protocol.
- If patient has any fluid collection, monitor MTX levels until MTX is below level of detection x 2 days.
## Increased MTX Concentrations and Leucovorin Rescue: St Jude OS2008

<table>
<thead>
<tr>
<th>MTX Level</th>
<th>Thresholds for action</th>
<th>Recommended LV rescue</th>
<th>IV fluids (mL/m²/hr)</th>
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<tbody>
<tr>
<td>24 hr</td>
<td>&lt; 10 µM</td>
<td>Protocol leucovorin rescue</td>
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<td>100 mg/m² IV q 6 hrs – start at hr 30</td>
<td>200</td>
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<td>250 mg/m² IV q 6 hrs – start immediately, admit patient</td>
<td>200</td>
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<tr>
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<td>500 mg/m² IV q 6 hrs – start immediately, admit patient</td>
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<tr>
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<td>&gt; 50 µM</td>
<td>Individualized, check that hydration and alkalization are adequate, check for nephrotoxic drugs, check creatinine, consider glucarpidase</td>
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How long to continue Leucovorin?

- Initiate within first 48 hrs
- Until MTX conc < 0.01 µM in patients at high risk for toxicity
High Risk Factors for Close Therapeutic Monitoring of MTX

- Ascites/pleural effusions – third spacing
- Poor renal function
  - Concurrent therapy with nephrotoxic agents
- Patient with emesis during infusion
- Patient with delayed clearance during previous course
Patient Case- HDMTX Monitoring

- AS is a 17-year old male with osteosarcoma. He is receiving his 3rd course of HDMTX (12 g/m²). He has previously received 2 courses of cisplatin (120 mg/m² per course).

- End of infusion [MTX] = 1069 uM
- 24 hr [MTX] = 19.4 uM
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What do you do for AS?

- Begin leucovorin at 100 mg/m² IV Q6 hours
- Hydrate at 200 ml/hr/m²
- Continue to check urine pH until [MTX] < 1 uM
- Check [MTX] at 48 hrs and adjust leucovorin per guidelines
- Continue to monitor [MTX] until < 0.01 uM
Goal of clinical pharmacokinetics: Individualize medicines to maximize cures and minimize adverse effects.
Thank you INCA

And thank you to my family!
# Methotrexate Bioanalytical Methods

<table>
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<tr>
<th>Method</th>
<th>Sample Matrix</th>
<th>Lower Limit of Quant.</th>
<th>Sample Volume</th>
<th>Factors</th>
<th>Preference</th>
</tr>
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<tr>
<td>HPLC (or LC MS/MS)</td>
<td>Plasma/Urine/CSF</td>
<td>0.01 μM</td>
<td>100 μl</td>
<td>Start-up costs; specialized training</td>
<td>Research settings; existing instrument</td>
</tr>
<tr>
<td>Immune-based</td>
<td>Plasma/Urine/CSF</td>
<td>0.05 μM</td>
<td>50-300 μl</td>
<td>High-throughput; cost-effective</td>
<td>Routine clinical use</td>
</tr>
<tr>
<td>Enzyme Inhibition</td>
<td>Plasma/Urine/CSF</td>
<td>0.02 μM</td>
<td>20-50 μl</td>
<td>Not in kit; interference</td>
<td>Not for routine use</td>
</tr>
</tbody>
</table>
Immune-Based Methods

- FPIA
  - TDxFLx

- EIA
  - Emit

- Chemiluminescent microparticle immunoassay
  - Abbott ARCHITECT
  - Available Late 2011
ARCHITECT Assay Format
Competitive 1-Step Chemiluminescence

Goat anti-mouse uparticle + Mouse anti-MTX MAb + Methotrexate-SPSP + MTX in Patient Sample

Wash, pretrigger, trigger, read
Correlation of MTX:
TDx vs. Architect (i1000)
## Cross-Reactivity and Interference Testing

<table>
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<th>Substance</th>
<th>Interference</th>
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</thead>
<tbody>
<tr>
<td>7-OH Methotrexate (1000 μM)</td>
<td>&lt;0.0025%</td>
</tr>
<tr>
<td>Folic acid (1000 μM)</td>
<td>&lt;0.002%</td>
</tr>
<tr>
<td>DAMPA (1000 μM)</td>
<td>&lt;28% (vs. 59% TDx)</td>
</tr>
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</table>

**Molecules:**
- **7-Hydroxy Methotrexate**
- **Folic Acid**
- **DAMPA**
Conversion from TDx Platform to Architect Late 2011

- Automated immuno-based assay method advantageous for our clinical practice

- ARCHECT i1000 results consistent with previous TDxFLx instrument with excellent performance characteristics (cross-reactivity, etc)

- Migrate clinical methotrexate assay to ARCHITECT i1000 by late 2011