Triazole Antifungal Therapeutic Drug Monitoring

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ECIL 6 meeting
September 11-12, 2015
Sophia Antipolis, France
Previous ECIL recommendations:
ECIL-4 2011: Pediatric fungal diseases
ECIL-5 2013: Antifungal recommendations

- TDM was not extensively addressed for antifungal treatment
- TDM was recommended for primary antifungal prophylaxis*

- Voriconazole → to improve efficacy, safety
  - Target trough: 1-5 mg/L
- Posaconazole → to improve efficacy
  - Target trough: > 0.7 mg/L
- Itraconazole → to improve efficacy, safety
  - Target trough: > 0.5 mg/L; toxicity 17.0 mg/L (bioassay)

*No evidence grading was applied in this section

ECIL 6 charges:

- Identify key questions concerning azole therapeutic drug monitoring (TDM) in patients with haematological malignancies / allogeneic HSCT
- Provide evidence-based recommendations or expert opinion addressing key questions (ESCMID/EFISG scoring system)

**Pharmacology**
- Russell Lewis (Italy)
- Roger Brüggemann (Netherlands)
- Christophe Padoin (France)

**Haematology / Infectious diseases**
- Johan Maertens (Belgium)
- Oscar Marchetti (Switzerland)
- Andreas Groll (Germany)

**Clinical reference laboratory**
- Elizabeth Johnson (UK)
- Maiken Arendrup (Denmark)
# Evidence grading-
ESC MID/EFISG scoring system

<table>
<thead>
<tr>
<th>Strength of Recommendation (SoR)</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grade A</td>
<td>ECIL strongly supports a recommendation for use</td>
</tr>
<tr>
<td>Grade B</td>
<td>ECIL moderately supports a recommendation for use</td>
</tr>
<tr>
<td>Grade C</td>
<td>ECIL marginally supports a recommendation for use</td>
</tr>
<tr>
<td>Grade D</td>
<td>ECIL supports a recommendation against use</td>
</tr>
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</table>

<table>
<thead>
<tr>
<th>Quality of Evidence (QoE)</th>
<th>Definition</th>
</tr>
</thead>
<tbody>
<tr>
<td>Level I</td>
<td>Evidence from at least 1 properly* designed randomized, controlled trial (orientated on the primary endpoint of the trial)</td>
</tr>
<tr>
<td>Level II</td>
<td>Evidence from at least 1 well-designed clinical trial (including secondary endpoints), without randomization; from cohort or case-controlled analytic studies (preferably from &gt; 1 centre; from multiple time series; or from dramatic results of uncontrolled experiments)</td>
</tr>
<tr>
<td>Level III</td>
<td>Evidence from opinions of respected authorities, based on clinical experience, descriptive case studies, or reports of expert committees</td>
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</table>

<table>
<thead>
<tr>
<th>Added Index</th>
<th>Source of Level II Evidence</th>
</tr>
</thead>
<tbody>
<tr>
<td>r</td>
<td>Meta-analysis or systematic review of RCT</td>
</tr>
<tr>
<td>t</td>
<td>Transferred evidence, that is, results from different patients’ cohorts, or similar immune-status situation</td>
</tr>
<tr>
<td>h</td>
<td>Comparator group: historical control</td>
</tr>
<tr>
<td>u</td>
<td>Uncontrolled trials</td>
</tr>
<tr>
<td>a</td>
<td>Published abstract presented at an international symposium or meeting</td>
</tr>
</tbody>
</table>

*poor quality of planning, inconsistency of results, indirectness of evidence etc... would lower the SoR
Key questions-Pharmacology

• What are the specificazole PK/PD considerations that support the need for TDM?

• Which triazoles should be monitored?
<table>
<thead>
<tr>
<th>Triazole</th>
<th>Significant PK variability?</th>
<th>Therapeutic range defined in humans?</th>
<th>Narrow therapeutic window?</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fluconazole</td>
<td>✔ yes</td>
<td>✔ yes</td>
<td>✗</td>
</tr>
</tbody>
</table>
TDM needed for fluconazole?

• Substantial PK variability (>30% CV) in some populations may result in subtherapeutic exposures
  • Critically-ill patients with sepsis\textsuperscript{1,2}
  • Hemodialysis (CVVH or CVVHD, CVVHDF)
  • Pediatrics\textsuperscript{3}

• Fluconazole has a broad therapeutic index \(\rightarrow\) possibly more practical to empirically administer higher weight-based doses (e.g., 8-12 mg/kg/day)

• Uncertainty regarding best monitoring strategy:
  • Estimate AUC/MIC (target > 100)...1, 2 and 4hr sample?

\begin{itemize}
\item \text{continuous venovenous hemofiltration (CVVH)}
\item \text{continuous venovenous hemodialysis (CVVHD)}
\item \text{continuous venovenous hemodiafiltration (CVVHDF)}
\end{itemize}

\textsuperscript{1,2} Ashbee et al. J Antimicrob Chemother 2014;69:1162-76.
\textsuperscript{3} Sinnollareddy et al. Crit Care 2015;19:33
\textsuperscript{3} van der Elst et al. Clin Infect Dis 2014;59:1527-1533
TDM may be beneficial for fluconazole in special circumstances?

- Hemodialysis/hemofiltration +/- sepsis
- CNS infections
- Pediatrics
- Infections cause by pathogens with elevated MICs (>2-4 mg/L)
- Patients at risk for QTc prolongation (especially in setting of renal disease)?

Fluconazole TDM may be helpful to guide dosing for rare treatment circumstances to target:
AUC/MIC > 100; AUC 400 mg·h/L; or trough of > 10 to 15 mg/L (BIII)
<table>
<thead>
<tr>
<th>Parameter</th>
<th>Substantial PK variability?</th>
<th>Therapeutic range defined in humans?</th>
<th>Narrow therapeutic window?</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fluconazole</td>
<td>✔ yes</td>
<td>✔ yes</td>
<td>✗ no</td>
</tr>
<tr>
<td>Itraconazole</td>
<td>✔ yes</td>
<td>✔ yes</td>
<td>✔ yes</td>
</tr>
<tr>
<td>Voriconazole</td>
<td>✔ yes</td>
<td>✔ yes</td>
<td>✔ yes</td>
</tr>
<tr>
<td>Posaconazole</td>
<td>✔ yes</td>
<td>✔ yes</td>
<td>? not well defined</td>
</tr>
<tr>
<td>Isavuconazole</td>
<td>✔ yes</td>
<td>✗ no</td>
<td>? not well defined</td>
</tr>
</tbody>
</table>

*Limited data for new posaconazole formulations
**Limited data
Key questions-Pharmacology

• What are the specific azole PK/PD considerations that support the need for TDM?

• Which triazoles should be monitored?

• What target levels are recommended for each triazole?

• When shouldazole concentrations be evaluated and re-evaluated? How is dosing adjusted?
Limitations of an evidence-based triazole target ranges

• A proportion of TDM evidence is derived from single-centre, retrospective, and/or statistically underpowered studies
• Many studies do not provide 95% CI when describing concentration-effect or toxicity relationships
• Current evidence supports an approximate TDM target range to maximize efficacy, and in some cases, safety
  – Ultimate dosing target is dictated by clinical situation (prophylaxis vs. treatment, severity or duration of infection, level of immunosuppression, susceptibility of pathogen...etc.)
Pharmacology: What target levels are recommended?

Itraconazole-PK variability

• Oral bioavailability
  • Capsule has variable, pH-dependent oral bioavailability (55%) → must give with food
  • Solution (cyclodextran): 30% higher bioavailability vs. capsule- absorption is pH independent but reduced with food → increased GI adverse effects
  • Mucositis, diarrhea associated with decreased blood levels; compliance with solution challenging

• Substrate and inhibitor of CYP3A4
  • Saturable, non-linear elimination
  • Complex chemistry (4 cis isomers with different affinity for CYP 3A4, PgP)
  • Active metabolite (OH-itra), 1 to 1.59-fold higher conc. then itraconazole → impacts interpretation of bioassay (2-10x higher than HPLC measurement, depending on calibration standards used)

**Pharmacology:** What target levels are recommended?

### Itraconazole concentration-efficacy relationship

- **Prophylaxis in neutropenic patients or other underlying conditions:** Breakthrough fungal infections are more frequent when trough itraconazole plasma levels < 0.25-0.5 mg/L (HPLC assay)\(^1\)-\(^4\)

- **Aspergillosis treatment:** improved outcomes with mean itraconazole plasma concentration of approximately 5-8 mg/L (bioassay) \(^5\)

- **Meta-analysis of 3,957 patients:** significant relationship between itraconazole dose and incidence of breakthrough IFI \(^6\)

**Efficacy target:**

- **Prophylaxis:** > 0.5 mg/L (parent compound only, HPLC assay method) **(AII)**
- **Treatment:** > 1 mg/L (parent compound only, HPLC assay method) **(AII)**

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Pharmacology: What target levels are recommended?

**Itraconazole concentration-toxicity relationship**

- Decreased rates of toxicity (fluid retention and GI adverse effects) at concentrations < 17 mg/L (bioassay) \(^1\)
  - CART analysis: 86% vs. 31%
- Estimation of HPLC safety target: < 3-4 mg/L (~ 5 fold lower than bioassay)\(^3\)

**Safety target:**
Prophylaxis and treatment (HPLC, parent compound): < 4 mg/L (BIII)
Bioassay method : < 17 mg/L (BII)

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Pharmacology: When should concentrations be evaluated?

Itraconazole-TDM approach

- **Itraconazole concentrations reach steady state after 2 weeks of therapy (if no loading doses administered)**\(^1,2\)
- **Check first trough level on day 5-7 or soon thereafter**
  - Earlier determinations may be indicated in the treatment of active disease: target > 0.25 mg/L
  - Due to long half-life, concentrations drawn in middle of dosing interval should not differ substantially from trough (essentially no plasma half-life)
- **Recheck trough sample in 7 days if:**
  - Changes affecting oral absorption
  - Change in dose
  - New interacting drug is started or stopped
  - Changes in clinical condition of patient

Pharmacology: How should doses be adjusted based on TDM results?

Itraconazole-TDM approach

- **If trough concentration is low (< 0.5 mg/L):**
  - Consider clinical scenarios that could be addressed to improve bioavailability (i.e. drug interactions, compliance, poor GI function, gastric pH)
  - Stop protein pump inhibitors, administer with Cola or other acidic beverage
  - Switch patient to oral solution or IV formulation if taking capsules
  - If capsules continued → increase dose by 100 mg twice daily
  - See specific recommendations for pediatric dosing

- **If trough concentration is high (> 4 mg/L by HPLC, > 17 mg/L bioassay assay method):**
  - Consider dose reduction if patient experiencing adverse effects or patient cannot be switched to alternative antifungal
**Pharmacology:** What target levels are recommended?

## Voriconazole PK variability

- **Bioavailability** 85-92% in healthy volunteers, but can be reduced (60-65%) in some populations, including pediatrics \(^4,6\)
  - Co-administration with food decreases absorption (AUC ↓ 35%)
- **Metabolism/clearance pathways associated with up to 100-fold intrapatient PK variability**
  - Patient CYP2C19 metabolic capacity (pharmacogenetics)\(^1,2\)
  - Non-linear saturable elimination in adults, changing metabolism rates (autoinduction)\(^2\)
  - Children < 12 years: 3-5 fold greater rate of CYP 2C19 metabolism. Adolescent clearance at ages 12-14 years depends on weight (50 kg)\(^5-10\)
  - Drug interactions- Substrate of CYP2C19, inhibitor of CYP3A4 \(^7\)
- **Little or no correlation between voriconazole dose and measured plasma level** \(^11-12\)

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**Pharmacology:** What target levels are recommended?

**Voriconazole concentration-efficacy relationship**

- Retrospective studies have identified a relationship between voriconazole trough concentrations in adult and paediatric patients and clinical outcome during prophylaxis or treatment.

- Some retrospective studies did not identify a relationship

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Pharmacology: What target levels are recommended?

Voriconazole concentration-efficacy relationship

1. **Prospective** studies have reported trough concentrations of ≥ 1.5-2 mg/L are associated with near maximal clinical response in treatment of IFI

2. **Post-hoc analysis of Phase II/III clinical trials:**
   - Vori C\textsubscript{avg} /MIC target > 2, or vori plasma 2-5 mg/L
   - Response rate: 74%

**Recommendation:** voriconazole prophylaxis and treatment target: > 1-2 mg/L *(AII)*;

higher troughs (> 2) are recommended for severe infections or when there are concern of treating fungi with elevated MICs

Pharmacology: What target levels are recommended?

**Voriconazole concentration-toxicity relationship**

- Patients with voriconazole trough concentrations > 5-6 mg/L have a higher probability of neurotoxic events and visual hallucinations; which may lead to premature discontinuation or interruption of therapy, and worse treatment outcome\(^1\-^4\)

- **Post-hoc phase II/III safety data analysis:**\(^5\)
  - Some evidence of relationship between increased risk of hepatotoxicity at higher voriconazole exposures
  - No reliable upper “cut-off” concentration can be identified to minimize risk of hepatotoxic effects\(^1,^5\)
  - Possible exception: Japanese patients hepatotoxicity was more common (34.5%) if voriconazole trough concentrations ≥ 3.9 mg/L\(^6\-^8\)

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Pharmacology: What target levels are recommended?

**Voriconazole-hepatotoxicity**

- Voriconazole plasma trough levels are not predictive of hepatotoxicity in Caucasian patients... 1-5
- However, voriconazole levels can be elevated in patients with liver dysfunction 7
- Oral therapy may be more hepatotoxic than IV due to first-pass effect 8,9
- CYP2C19 genotype not independently associated with hepatotoxicity risk
- In Japanese patients hepatotoxicity was more common (34.5%) if voriconazole trough concentrations ≥ 3.9 mg/L 10-12
  - CYP2C19 HET or HOM poor metaboliser genotype frequency 60-70%:
  - Proposed therapeutic range 2-4 mg/L

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Voriconazole plasma concentrations did not predict hepatotoxicity in Phase II/III clinical trials
Voriconazole plasma concentrations are associated with clinical response and neurotoxicity

Clinical response, IFI

Neurotoxic AE, IFI
**Pharmacology:** What target levels are recommended?

Voriconazole concentration-toxicity relationship

**Recommendation:** voriconazole safety target: < 5.0-6.0 mg/L (AII);

Patients without symptoms of clinical toxicity may not require dose reductions, however the risk versus benefit must be weighed for each patient.

Maintenance of exposures near this threshold may be needed for severe infections (e.g., CNS infection) or when treating fungi with elevated MICs.

Lower trough < 4 mg/L in Japanese patients may be associated with lower hepatotoxicity risk (CYP2C19 genotype/higher exposures).
Pharmacology: When should concentrations be evaluated?

**Voriconazole TDM approach**

First trough sample 2-5 days (or after 5th dose including loading doses):

Trough should be repeated during second week of therapy to confirm patient in therapeutic range (1-6 mg/L):

Recheck trough 3-5 days if:

- Change in dose
- IV to oral switch
- Change in clinical condition (e.g., uncontrolled IFI or suspected toxicity)
- New interacting drug is started or stopped
Pharmacology: How should doses be adjusted based on TDM results?

Voriconazole TDM approach

- **If pre-dose trough concentration is low (< 1 mg/L)**
  - Check to ensure if dose was adequate (including loading dose)
  - Screen for clinical scenarios affecting voriconazole PK (e.g., compliance, drug interactions)
  - If recently switched from IV to oral, administer same weight-based (mg/kg) oral dose
    - lower levels often associated with fixed 200 mg BID oral dose
  - If receiving oral therapy, consider switch to IV
  - If plasma levels are very low (< 0.5 mg/L), consider dose IV or oral dose increase daily dose by 50%. Adjust subsequent doses based on TDM results *(see nomogram on slide # 28)*
  - If receiving IV therapy, increase daily dose *(see nomogram on slide # 28)* and recheck plasma level after 2-5 days.

- Computerized dosing assistance programs: e.g., DoseMe®, Insight Rx® can aid dosage selection and probability of target attainment
- See specific slides for recommended pediatric dosing
**Pharmacology:** How should doses be adjusted based on TDM results?

**Voriconazole TDM approach**

- If pre-dose trough concentration is high (> 6 mg/L)
  - Double check the sample is indeed a pre- and not post- dose sample
  - Screen for clinical scenarios affecting voriconazole PK (e.g., drug interactions, appropriate dose per weight)
  - Dose reduction may not be necessary if patient is tolerating voriconazole- However, the risk versus benefit is a decision individualised for each patient
  - CYP2C19 genotyping not currently recommended for patients monitored with routine TDM
  - Dose reduction protocol (Park et al CID 2012):¹
    - Reduced drug discontinuation, but **not** adverse effects
    - Reduce dose by 50% if level elevated, no adverse effect
    - If adverse effect and elevated level, or trough > 10 mg/L: hold one dose and reduce subsequent doses by 50%
    - Alternative approach: Dose by TDM results and nomogram (see slide #30)
    - Computerized dosing assistance programs may be helpful: e.g., DoseMe®, Insight Rx®

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**Pharmacology:** How should doses be adjusted based on TDM results?

**Voriconazole TDM-guided dosing algorithm**

<table>
<thead>
<tr>
<th>Voriconazole trough level</th>
<th>Dosage each 12 hrs (oral)</th>
<th>Dosage each 12 hrs (intravenous)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;0.1mg/L</td>
<td>400mg* 250mg* 300mg* 400mg*</td>
<td>4mg/kg* 5mg/kg* 6mg/kg* 7mg/kg*</td>
</tr>
<tr>
<td>0.1-0.4mg/L</td>
<td>400mg 400mg 400mg 500mg</td>
<td>6mg/kg 7mg/kg 8mg/kg 8.5mg/kg</td>
</tr>
<tr>
<td>0.5-1mg/L</td>
<td>300mg 300mg 400mg 450mg</td>
<td>5mg/kg 6mg/kg 7mg/kg 8mg/kg</td>
</tr>
<tr>
<td>1-1.5mg/L</td>
<td>250mg 300mg 450mg 450mg</td>
<td>5mg/kg 6mg/kg 7mg/kg 8mg/kg</td>
</tr>
<tr>
<td>1.5-2mg/L</td>
<td>250mg 300mg 450mg 450mg</td>
<td>5mg/kg 6mg/kg 7mg/kg 8mg/kg</td>
</tr>
<tr>
<td>2-3.5mg/L</td>
<td>200mg 250mg 300mg 400mg</td>
<td>4.5mg/kg 5.5mg/kg 6.5mg/kg 7.5mg/kg</td>
</tr>
<tr>
<td>3.5-5mg/L</td>
<td>150mg 200mg 250mg 300mg</td>
<td>4mg/kg 5mg/kg 5mg/kg 6mg/kg</td>
</tr>
<tr>
<td>&gt; 5mg/L</td>
<td>100mg 150mg 150mg 200mg</td>
<td>3mg/kg 4mg/kg 4mg/kg 5mg/kg</td>
</tr>
</tbody>
</table>

* = dosage given to patient at time of concentration measurement

Slide courtesy of Roger Brüggemann
**Pharmacology:** What target levels are recommended?

**Posaconazole PK variability**

- **Oral bioavailability (suspension)**
  - Affected by gastric pH, frequency of dosing, and administration with (fatty) food
  - Decreased when administered with proton pump inhibitors
  - Decreased by GI disease (diarrhea, mucositis)
  - Decreased absorption when administered by NG tube
- **PK problems in past compounded by lack of IV formulation**

33% higher bioavailability in fasted subjects versus suspension in fed subjects

Absorption of new tablet formulation does not depend on low gastric pH, and less affected by food → preferred oral formulation if patients can take tablets *(AII)*

3-Dolton et al. Antimicrob Agent Chemother 2014;58:6879-6885
Pharmacology: What target levels are recommended?

Posaconazole PK variability

- **Distribution:**
  - Large Vd, highly protein bound (> 98%, mostly albumin)
  - Vd increased in neutropenic patients, during active fungal disease vs. healthy volunteers¹
- **Metabolism/Clearance:**
  - Hepatic metabolism by UDP pathway to a monoglucuronide of posaconazole (18 - 28% of profiled radioactive dose). Only minor metabolites are formed by CYP450-mediated pathways
  - **Non-linear clearance observed with escalating IV doses**²

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1-Dolton et al. Antimicrob Agent Chemother 2014;58:6879-6885
Pharmacology: What target levels are recommended?

**Posaconazole concentration- prophylaxis efficacy**

- **Pharmacokinetic analysis of two pivotal prophylaxis trials utilizing suspension formulation did not report significant concentration-effect relationships**
  - Median posaconazole 0.61 mg/L (breakthrough IFI) vs. 0.92 mg/L (no breakthrough)

- **FDA pharmacodynamic analysis:**
  - Inverse relationship between POS plasma levels and clinical failure by logistic regression
  - Proposed efficacy target: 0.7 mg/L
  - Definition of clinical failure used in this analysis was different than original studies (resulted in a greater number of treatment failures)

Pharmacology: What target levels are recommended?

Posaconazole concentration- prophylaxis efficacy

- Other monocentric studies reported concentration-response relationship between posaconazole plasma trough levels and risk of breakthrough infection\(^1-^5\)
  - > 0.5 or 0.7 mg/L

**Recommendation: prophylaxis target: > 0.7 mg/L (BII)**

**Tablet formulation (or IV formulation) are preferred formulations to maximize probability of achieving target plasma levels (AII)**

Pharmacology: What target levels are recommended?

Posaconazole concentration – treatment efficacy

- Open-label salvage study of posaconazole salvage therapy in patients with invasive aspergillosis refractory or intolerant to other antifungals¹

<table>
<thead>
<tr>
<th>Patient Quartile</th>
<th>Cavg Range mg/L</th>
<th>Clinical Failure</th>
</tr>
</thead>
<tbody>
<tr>
<td>Q1</td>
<td>0.055 – 0.277</td>
<td>76%</td>
</tr>
<tr>
<td>Q2</td>
<td>0.290 – 0.544</td>
<td>47%</td>
</tr>
<tr>
<td>Q3</td>
<td>0.550 – 0.861</td>
<td>47%</td>
</tr>
<tr>
<td>Q4</td>
<td>0.877 – 2.010</td>
<td>29%</td>
</tr>
<tr>
<td>Hist. Control</td>
<td>--</td>
<td>74%</td>
</tr>
</tbody>
</table>

Recommendation: treatment efficacy trough > 1 mg/L (AII) (defined for invasive aspergillosis)
Pharmacology: What target levels are recommended?

Posaconazole concentration- toxicity

- No relationship between adverse effects and plasma concentrations for oral suspension \(^1\text{-}^3\)
- Pharmacokinetic bridging studies for gastroresistant tablet and IV formulation used an upper plasma target of 3.75 mg/L\(^3\)

Recommendation: At present, insufficient data to recommend target trough for safety *further data are needed*

**Pharmacology:** When should concentrations be evaluated?

### Posaconazole gastroresistant tablet and IV formulations

Up to 10% of patients receiving new posaconazole formulations may not achieve plasma targets > 0.7 mg/L. The percentage of patients not reaching treatment target (> 1 mg/L) will be higher.

It is unknown whether risk for inadequate exposures can be predicted based on observable clinical risk factors alone (e.g., mucositis, aGVHD). Therefore, TDM remains the most direct approach for identifying patients with suboptimal posaconazole plasma levels.

- Pending further data, TDM is still recommended in patients receiving posaconazole tablets or IV formulation for prophylaxis (**CIII**)
- TDM is recommended in patients receiving posaconazole tablets or IV formulation receiving treatment for suspected or documented fungal infection (**CIII**)
- TDM is indicated for patients receiving tablets or IV formulation in the setting of breakthrough or progressing infection unresponsive to treatment, treatment of pathogens with reduced susceptibility, or drug interactions (**CIII**)

*additional data are needed*

Pharmacology: How should doses be adjusted based on TDM results?

Posaconazole TDM approach

- If pre-dose trough concentration is low (< 0.7 mg/L prophylaxis or < 1 mg/L treatment):
  - Assess clinical scenarios affecting bioavailability and compliance.
  - Switch patient to the gastro-resistant tablet or IV formulation if receiving suspension
  - If patient requires suspension formulation, increase dose from 600 to 800 mg daily administered in 4 divided doses with food or acidic beverage, stop acid suppression therapy if feasible
  - Recheck trough after 5-7 days
  - Safety of dose escalation with gastro-resistant tablets above 300 mg day is not well defined

Isavuconazole-PK variability

**Pharmacology:** What target levels are recommended?

### Absorption
- Administered as prodrug (isavuconazolium sulfate)
- 98% bioavailability, not affected by food or gastric pH

### Distribution
- Vd 450 L (high tissue distribution)
- Requires loading dose 200 mg q8h x 48h then 200 mg daily

### Metabolism
- Metabolized via CYP3A4 → UGT
- Moderate inhibitor of CYP3A4
- Very long half-life (60-130 hours, increased in hepatic impairment)
- Less pharmacokinetic variability versus voriconazole

**Isavuconazole**
(active drug BAL 4815)

\[
\text{logP 3.92}
\]
\[
pKa 2.70
\]
**Pharmacology:** What target levels are recommended?

**Isavuconazole-concentration efficacy**

Isavuconazole package labelling:

12.2 Pharmacodynamics

**Pharmacokinetic/Pharmacodynamic Relationship**
In patients treated with CRESEMBA for invasive aspergillosis in a controlled trial, there was no significant association between plasma AUC or plasma isavuconazole concentration and efficacy.

TDM is indicated for patients receiving tablets or IV formulation in the setting of breakthrough or infection unresponsive to treatment, treatment of pathogens with reduced susceptibility, or in the setting of drug interactions (CIII)

*additional data are needed*
### Summary of TDM plasma target level recommendations

<table>
<thead>
<tr>
<th>Triazole</th>
<th>Recommended plasma rangea</th>
<th>SOR</th>
<th>Timing of first trough sample</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Voriconazole</strong></td>
<td>Prophylaxis and treatment:</td>
<td>All (efficacy)</td>
<td>After 2-5 days; (repeat sampling recommended)</td>
</tr>
<tr>
<td></td>
<td>Acceptable: 1-6 mg/L;</td>
<td>All (toxicity)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Optimal: 2-5 mg/L</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Posaconazole</strong></td>
<td>Prophylaxis: &gt; 0.7 mg/L</td>
<td>BII (efficacy)</td>
<td>Tablet/IV: after 3 days:</td>
</tr>
<tr>
<td></td>
<td>Treatment: &gt; 1.0 mg/L</td>
<td>All (efficacy)</td>
<td>Suspension: 5-7 days.*</td>
</tr>
<tr>
<td><strong>Itraconazole</strong></td>
<td>Prophylaxis: 0.5-4 mg/L</td>
<td>All (efficacy)</td>
<td>7-15 days;*</td>
</tr>
<tr>
<td></td>
<td>Treatment: 1-4 mg/L</td>
<td>BII (toxicity)</td>
<td></td>
</tr>
</tbody>
</table>

*a values from a chromatography assay: i.e. high performance liquid chromatography (HPLC), liquid chromatography mass spectroscopy (LC/MS) of LC/MS/MS

b patients without symptoms of clinical toxicity may not warrent dosage adjustment, decisions should be individualised to the patient

c higher troughs (≥ 2) are advocated for severe infections or treatment of pathogens with potentially or documented elevated MICs (around 1 mg/L or higher)

*earlier sampling possible and may be desirable during treatment.

* Earlier sampling possible using lower targets
# Recommended prophylaxis plasma target ranges - Guideline comparisons

<table>
<thead>
<tr>
<th></th>
<th>Fluconazole (mg/L)</th>
<th>Itraconazole (mg/L)</th>
<th>Voriconazole (mg/L)</th>
<th>Posaconazole (mg/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>ECIL-6</strong></td>
<td>TDM not routinely recommended</td>
<td>0.5-4 (HPLC)</td>
<td>1-6</td>
<td>&gt; 0.7</td>
</tr>
<tr>
<td><strong>Ashbee et al. 2014</strong></td>
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<td>0.5-4 (HPLC)</td>
<td>1-6</td>
<td>&gt; 0.7</td>
</tr>
<tr>
<td><strong>Hamada et al. 2013 (VOR specific)</strong></td>
<td>TDM not routinely recommended</td>
<td>--</td>
<td>1-5</td>
<td>--</td>
</tr>
<tr>
<td><strong>Scodavolpe et al. 2014</strong></td>
<td>AUC/MIC &gt; 25</td>
<td>&gt; 0.5 (HPLC, MIC dependent)</td>
<td>1-6</td>
<td>&gt; 0.5</td>
</tr>
<tr>
<td><strong>Chau et al. 2014</strong></td>
<td>TDM not routinely recommended</td>
<td>&gt; 0.5-1 (HPLC, MIC dependent)</td>
<td>1-6</td>
<td>&gt; 0.7</td>
</tr>
</tbody>
</table>

## Recommended treatment plasma target ranges - Guideline comparisons

<table>
<thead>
<tr>
<th></th>
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<td>&gt; 1</td>
</tr>
<tr>
<td>Ashbee et al. 2014</td>
<td>TDM not routinely recommended</td>
<td>1-4 (HPLC) recommended higher MIC</td>
<td>1-6</td>
<td>&gt; 1</td>
</tr>
<tr>
<td>Hamada et al. 2013. (vori specific)</td>
<td>--</td>
<td>--</td>
<td>1-5</td>
<td>--</td>
</tr>
<tr>
<td>Scodavolpe et al. 2014</td>
<td>AUC/MIC &gt; 25</td>
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<td>1-5</td>
<td>0.5-1.5</td>
</tr>
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<td>&gt; 0.5-1 (HPLC)</td>
<td>1-6</td>
<td>&gt; 1</td>
</tr>
</tbody>
</table>

Key questions-Pharmacology

• What are the specific azole PK/PD considerations that support the need for TDM?
• Which triazoles should be monitored?
• What target levels are recommended for each triazole?
• When should azole concentrations be evaluated and re-evaluated? How is dosing adjusted?
• What is the role for TDM in managing drug interactions?
Key questions - pharmacology: What is the role of TDM for drug interactions?

**Drug interactions affecting azole levels**

- **Patient receiving co-medication that induces CYP-P450 enzymes:**
  - Change in therapy to non-interacting antifungal recommended *(AII)*
- **Patient receiving co-medication that induces UGT enzymes:**
  - TDM recommended for posaconazole *(AII)*
- **Patient receiving antacids and PPI with itraconazole capsules or posaconazole suspension**
  - TDM recommended *(AII)*
Key questions-pharmacology: What is the role of TDM for drug interactions?

Azole affects on metabolism of other drugs

- Patients should have medication records screened using suitable computerized screening database before starting and stopping antifungals (AIII)
  - Examples: [www.fungalpharmacology.org](http://www.fungalpharmacology.org); [www.aspergillus.ork.uk/content/antifungals-drug-interactions](http://www.aspergillus.ork.uk/content/antifungals-drug-interactions), or commercial products such as Lexi-comp Lexi Interact®
- Patient receiving co-medication metabolized through CYP P450 → esp. CYP3A4:
  - Consult drug interactions database or clinical pharmacologist (AIII)
- Medications inducing UGT enzymes
  - Consult drug interactions database or clinical pharmacologist (AIII)
Key questions - Haematology / Infectious diseases

• What clinical scenarios in patients with haematological malignancies or HSCT receiving triazoles benefit from TDM assessment?

• How should TDM be used to optimize triazole use in paediatric patients with haematological malignancy or HSCT?

• Who should advise, interpret and follow-up on TDM results?
<table>
<thead>
<tr>
<th>Scenarios where routine azole TDM should be considered</th>
<th>Examples, comment</th>
<th>SOR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Populations with diseases or underlying risk factors for pharmacokinetic variability</td>
<td>Impaired GI function; hepatic dysfunction (voriconazole, posaconazole, itraconazole, isavuconazole); pediatric patients, elderly patients, obese patients, malnourished, malignancy-associated cachexia, critically-ill patients; Intravenous to oral switch, changing GI function, changing hepatic function, physiological-instability</td>
<td>All</td>
</tr>
<tr>
<td>Interacting medications that could reduce or increase triazole clearance</td>
<td>Patient receiving medication that induces CYP3A4 (antiretroviral medications, anti-epileptic, or rifamycins), antacids, proton-pump inhibitors (itraconazole capsules, posaconazole suspension)</td>
<td>All</td>
</tr>
</tbody>
</table>
### Haematology & ID: Which clinical scenarios will benefit from TDM?

<table>
<thead>
<tr>
<th>Scenarios where azole TDM is likely to be useful in patients with hematologic malignancies</th>
<th>Examples, comment</th>
<th>SOR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Severe infections</td>
<td>Extensive or bulky infection, lesions contiguous with critical structures, CNS infection, multifocal or disseminated infection</td>
<td>AII</td>
</tr>
<tr>
<td>Compliance</td>
<td>Important issue with longer-term consolidation therapy or secondary prophylaxis (outpatient)</td>
<td>AII</td>
</tr>
<tr>
<td>Suspected breakthrough infection</td>
<td>TDM can establish whether fungal disease progression occurred in the setting of inadequate antifungal exposure</td>
<td>AII</td>
</tr>
<tr>
<td>Suspected drug toxicity, especially neurotoxicity (voriconazole)</td>
<td>Although exposure-response relationships are described for other toxicities (e.g., hepatotoxicity), the utility of TDM to prevent their occurrence is less well established</td>
<td>AII</td>
</tr>
</tbody>
</table>
**Haematology & ID:** Which clinical scenarios will benefit from TDM?

<table>
<thead>
<tr>
<th>Scenarios where azole TDM is likely to be useful in patients with hematologic malignancies</th>
<th>Examples, comment</th>
<th>SOR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment of a pathogen with reduced susceptibility</td>
<td>Consequences of pharmacokinetic variability are more severe with increasing MIC</td>
<td>All</td>
</tr>
</tbody>
</table>

Key questions - Haematology / Infectious diseases

• What clinical scenarios in patients with haematological malignancies or HSCT receiving triazoles benefit from TDM assessment?
• How should TDM be used to optimize triazole use in paediatric patients with haematological malignancy or HSCT?
• Who should advise, interpret and follow-up on TDM results?
Fourth European Conference on Infections in Leukaemia (ECIL-4): guidelines for diagnosis, prevention, and treatment of invasive fungal diseases in paediatric patients with cancer or allogeneic haemopoietic stem-cell transplantation

Andreas H Groll, Elio Castagnolo, Simone Cesaro, Jean-Hugues Dalle, Dan Engelhard, William Hope, Emmanuel Roilides, Jan Styczynski, Adilia Warnis, Thomas Lehmbelder, on behalf of the Fourth European Conference on Infections in Leukaemia, a joint venture of the Infectious Diseases Working Party of the European Group for Blood and Marrow Transplantation (EBMT-IDWP), the Infectious Diseases Group of the European Organisation for Research and Treatment of Cancer (EORTC-IDG), the International Immunocompromised Host Society (IICHS), and the European Leukaemia Net (ELN)

**ECIL 4:** TDM included with all recommendations for use of voriconazole, posaconazole, and itraconazole in children

**ECIL 6:** TDM targets and approach harmonized with adult recommendations

Routine TDM is recommended in pediatric heme malignancy / HSCT patients treated with itraconazole, voriconazole or posaconazole (AII)
Background and Principles \(^1,^2\)

- Key issue relative to adults is different PK / dosing
  - \(\leq 12\) years: Greater clearance / larger doses
  - \(> 13\) years: PK / dosing mostly similar vs. adults

- Pharmacodynamics and PK/PD relationships can be considered similar in management of IFDs

- PK/safety studies in all pediatric age groups are prerequisite for safe and effective use
- No larger pediatric PK/PD studies required, adult data can be used to support PK/PD principles

2. European Medicines Agency. CPMP/ICH/2711/99
# Pharmacological considerations

<table>
<thead>
<tr>
<th>Antifungal Agent</th>
<th>Approved Indications *</th>
<th>Ped. Dosage Range **</th>
<th>Specific Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fluconazole</td>
<td>Treatment / prevention of superficial / invasive Candida infections; treatment of cryptococcosis and coccidioidomycosis</td>
<td>8-12 mg/kg/d IV/PO in one single dose; no routine TDM</td>
<td>Increased weight-normalized plasma clearance relative to adults; optimal dose uncertain. ECIL 4 recommends 8-12 mg/kg/d (max. 400 mg/d) for prophylaxis and 8-12 mg/kg/d (max. 800 mg/d) for targeted treatment. Recent retrospective PK/PD analyses suggest to use the maximum approved dose of 12 mg/kg/d (max. 800mg/d) for targeted treatment. Potential for drug-drug interactions.</td>
</tr>
<tr>
<td>Itraconazole</td>
<td>Treatment of superficial Candida infections; 2nd line treatment of invasive candidiasis, aspergillosis and cryptococcosis; prophylaxis in granulocytopenic patients</td>
<td>5 mg/kg/d PO in two divided doses plus TDM</td>
<td>Limited pediatric PK data in 2 to 17 year old subjects for oral suspension, no principal differences relative to adults. Similar problems with absorption. ECIL 4 recommends 5 mg/kg/d in two divided doses for prophylaxis and treatment. Only single dose PK data available for the IV formulation. Not licensed in the EU in subjects &lt;18 years, no PK data for children &lt;2 years. High potential for relevant drug-drug interactions.</td>
</tr>
<tr>
<td>Posaconazole</td>
<td>2nd line treatment of aspergillosis, fusariosis, chromoblasto- and coccidioidomycosis; treatment of oropharyngeal candidiasis; prophylaxis in AML/MDS and allogeneic HSCT patients</td>
<td>600-800 mg/d PO in 2 to 4 divided doses plus TDM</td>
<td>Limited pediatric PK data for the oral suspension; no principal differences relative to adults in adolescents ≥13 years. Similar problems with absorption. Not licensed in subjects &lt;18 years in the EU but licensed in adolescents ≥13 years in the US for prophylaxis. ECIL 4 recommends 600 mg/d in three divided doses for prophylaxis and 800 mg/d in 2 or 4 divided doses for treatment in subjects ≥13 years. No pediatric PK data exist for the novel tablet- and the IV formulation; however, PK (and dosing) in adolescents ≥13 years are expected to be similar relative to adults. High potential for relevant drug-drug interactions.</td>
</tr>
<tr>
<td>Voriconazole</td>
<td>Treatment of invasive aspergillosis, fusariosis, scedosporiosis; treatment of candidaemia in non-granulocytopenic patients; prophylaxis in allogeneic HSCT patients</td>
<td>2-12 yrs /12-14 yrs and &lt;50kg: 8 mg/kg BID (day 1: 9 mg/kg) IV and 9 mg/kg BID PO; ≥15 yrs and 12-14 yrs and ≥50kg: 4 mg/kg BID (day 1: 6 mg/kg) IV; 200 mg BID PO plus TDM (all)</td>
<td>Increased age-dependent weight-normalized plasma clearance and lower oral bioavailability relative to adults; similar, if not higher PK variability. Similar doses recommended for prophylaxis and treatment. Not licensed in subjects &lt;2 years of age. High potential for relevant drug-drug interactions and relevant potential for hepatic, cutaneous, and neurological adverse events</td>
</tr>
</tbody>
</table>

* Summarised; or specific wording, please refer to the summary of product characterics (SPCs); ** as recommended by ECIL 4

IV, intravenously; PO, orally; TDM, therapeutic drug monitoring; PK, pharmacokinetics; for references please refer to the appendix

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Haematology & ID: TDM in pediatric patients?

**Pediatric patients-ECIL 4 recommendations**

- **Voriconazole**
  - **Children 2-12 years > 50 kg:**
    - IV: 9 mg/kg twice daily day1, then 8 mg/kg twice daily
    - Oral: 9 mg/kg twice daily
  - **Children ≥ 15 years or 12-14 > 50 kg:**
    - Use adult dosing
  - **TDM is recommended, dosing target trough (same as adults):**
    - Efficacy > 1-2 mg/L
    - Safety < 5-6 mg/L
  - **Increased mortality OR 2.6 (1.4-4.8) if VRC < 1 mg/L**

**Routine TDM is recommended in pediatric cancer / HSCT patients treated with voriconazole (AII)**

Pediatric patients-ECIL 4 recommendations

- **Posaconazole prophylaxis**
  - No pediatric data on the tablet or IV formulations in pediatrics < 12 years; limited data for suspension (off-label)
    - **Children > 12 years**
      - 600 mg/d of the susp. in 3 divided doses with food
      - TDM is recommended, dosing target trough >0.7 mg/L
- **Posaconazole primary or salvage therapy**
  - **Children > 12 years**
    - 800 mg/d of the susp. in 2 or 4 divided doses with food
    - TDM is recommended, dosing target trough >1 mg/L

Routine TDM is recommended in paediatric haematology patients treated with posaconazole *(AII)*
Pediatric patients-ECIL 4 recommendations\(^1\)

- **Itraconazole prophylaxis**
  - **Children > 2 years**
    - 5 mg/kg/d of the suspension orally in two divided doses
    - TDM is recommended, dosing trough target > 0.5 mg/L

- **Itraconazole salvage treatment**
  - **Children > 2 years**
    - 5 mg/kg/d of the suspension orally in two divided doses
    - Consider loading dose 10 mg/kg/day (two divided doses days 1-2) in patients with severe disease
    - TDM is recommended, dosing trough target > 1 mg/L

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**Routine TDM is recommended in pediatric haematology patients treated with itraconazole** (AII)

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TDM-approach in pediatric patients

• Similar general principles/strategies as adults

• If dose adjustments are indicated:
  • In the absence of specific data, dose adjustments of at least 50% of the last total daily dose are recommended if plasma levels are low (posaconazole suspension: administer in 4 daily doses)
  • Recheck trough levels after 5 (voriconazole, posaconazole) to 7 (itraconazole) days (AII)
Haematology & ID: TDM in pediatric patients?

Key pediatric references

Fluconazole:

Itraconazole:
4. Foot AB, Veys PA, Gibson BE. Itraconazole oral solution as antifungal prophylaxis in children undergoing stem cell transplantation or intensive chemotherapy for haematological disorders. Bone Marrow Transplant 1999; 24: 1089–93
Key pediatric references

Voriconazole:
**Key pediatric references**


**Posaconazole:**


Key questions - Haematology / Infectious diseases

- What clinical scenarios in patients with haematological malignancies or HSCT receiving triazoles benefit from TDM assessment?
- How should TDM be used to optimize triazole use in paediatric patients with haematological malignancy or HSCT?
- Who should advise, interpret and follow-up on TDM results?
Haematology & ID: Who should advise, interpret, and follow-up on TDM results?

**Therapeutic drug monitoring process**

**Pre-analytical phase**
- Clinical question
- Test selected
- Test ordered
- Specimen collected

**Analytical phase**
- Sample prepared
- Analysis performed
- Results verified

**Post-analytical phase**
- Results reported
- Clinical answer
- Action taken
- Effect on patient care
TDM process recommendations

• Multidisciplinary approach to TDM is recommended
  • Should include involvement of nursing staff, physicians (haematologist and infectious diseases), analytical staff, pharmacologist, and microbiologist with clearly-defined responsibilities (AIII)

• Pre-analytical phase
  • Patient sampling schemes should be standardized when possible to minimize errors (AIII)
  • Trough concentrations are generally the least-error prone and most convenient approach to measure patient azole exposure

Haematology & ID: Who should advise, interpret, and follow-up on TDM results?

Haematology & ID: Who should advise, interpret, and follow-up on TDM results?

**TDM process recommendations**

- **Analytical phase**
  - An accurate, precise, sensitive, and selective analytical method for the quantitative determination of azole antifungal drugs in plasma/serum is recommended (AIII)
  - Assays should be validated according to the current requirements for validation of bioanalytical assays¹ (AIII)
  - To help identify sources of errors and to further improve analytical methods, participation in an ongoing proficiency testing program is recommended (AIII)
    - Standards and controls should be externally validated at a certified centre by HPLC

¹ EMA Guideline on bioanalytical method validation. 2011.
Haematology & ID: Who should advise, interpret, and follow-up on TDM results?

TDM process recommendations

• Post-analytical phase
  • Interpretation of results should be performed by clinical pharmacist/pharmacologist, physician or microbiologist with expertise in antifungal therapy familiar with the sampling time, patient clinical parameters, and likely pathogen if not identified (AIII)

• Results should be communicated with responsible physician by someone with expertise in TDM and interpretation (BIII)

• Repeat sampling should be considered once-or twice weekly in patient strongly suspected or proven to have invasive fungal disease or clinical instability, or concentration outside target range
  • Need for resampling is individualized to the clinical scenario o the patient

Key questions-laboratory

- What samples are suitable for analysis?
- How should antifungal drugs be analysed?
- What are the external quality assurance/assessment (EQA) schemes for laboratories analysing TDM samples?
Laboratory: What should be monitored

Measure parent drug only or parent + metabolites?

- **Itraconazole**
  - Parent drug
  - OH-itraconazole metabolite (active)
- **Voriconazole**
  - Parent drug
  - Optional: Voriconazole –N-oxide metabolite (inactive)
- **Posaconazole**
  - Parent drug only
- **Isavuconazole**
  - Parent drug only

When analysed with parent drug could provide information on compliance, metabolic phenotype...

Laboratory: How should antifungal drugs be analysed?

**Reporting antifungal drug levels:**

- Levels should be reported as mg/L to one decimal place for microbiological methods and two decimal places for instrumental methods.

- Very low levels may have to be reported as < whatever value has been obtained as the lower limit of detection for that method.

- Very high levels can be reported as > whatever value has been obtained as the upper limit of detection but, are more useful for dosage adjustment when diluted and repeated to calculate an absolute value.

- It is important to include an interpretation with the drug level i.e. low level, high level or satisfactory level and an indication of normal ranges and efficacy and toxicity cut offs if known.
What samples are suitable for analysis?

Sample types validated for separate or simultaneous TDM for itraconazole, posaconazole, voriconazole (+/- more antifungals) by HPLC/LC-MS

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>HPLC / LC-MS</th>
<th>References (examples)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plasma</td>
<td>yes</td>
<td>Decostard doi:10.1128/AAC.00404-10 Verweij-van Wissen</td>
</tr>
</tbody>
</table>
| CSF                       | yes          | Wiederhold doi:10.1128/AAC.01558-13 voriconazole (and some data also on posaconazole though less clinically relevant)
What samples are suitable for analysis?

Sample types validated for TDM for itraconazole, posaconazole, voriconazole (separately) by bioassay.

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>Bioassay</th>
<th>References (examples)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum</td>
<td>yes</td>
<td>Cendejas-Bueno doi:10.1128/AAC.00323-13 (voriconazole)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Odds doi: 10.1093/jac/43.5.723 (itraconazole)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Pascual doi:10.1128/AAC.00957-06 (voriconazole)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Pascual doi 10.1128/AAC.00022-10 (posaconazole)</td>
</tr>
<tr>
<td>Plasma</td>
<td>yes</td>
<td>Pascual doi:10.1128/AAC.00957-06 (voriconazole)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Pascual doi 10.1128/AAC.00022-10 (posaconazole)</td>
</tr>
<tr>
<td>Dried Blood Spot</td>
<td></td>
<td>No data</td>
</tr>
<tr>
<td>Dried Plasma Spot</td>
<td></td>
<td>No data</td>
</tr>
<tr>
<td>CSF</td>
<td></td>
<td>No data</td>
</tr>
</tbody>
</table>
**Laboratory:** How should antifungal drugs be analysed?

Two main types of analytical method for measuring antifungal drug levels:

1. **Microbiological method:** plate assay / bioassay
   - plate seeded with susceptible organism
   - known standard concentrations placed in triplicate wells / discs
   - patient samples placed in triplicate wells / discs
   - plate incubated and zones of inhibition measured
   - standard curve constructed to interpolate unknowns

2. **Instrumental techniques:** although there are a large number of potential electrophoretic and chromatographic methods for antifungal drug analysis high-performance liquid chromatography (HPLC), ultra-HPLC and HPLC with mass spectrophotometry (HPLC-MS) have become the reference methods.

   ARK Diagnostics, Inc. has developed an enzyme immunoassay test for Voriconazole TDM that can be run on various biochem lab robots like Roche’s Cobas 8000 instrument. (Cattoir et al Clin Chem Lab Med 2015; 53(5):e135-9)
## Laboratory: How should antifungal drugs be analysed?

<table>
<thead>
<tr>
<th>Method</th>
<th>Advantages</th>
<th>Disadvantages</th>
</tr>
</thead>
</table>
| Bioassay            | Inexpensive to conduct  
Acquisition costs minimal  
Minimal training  
Quick set up  
Suitable for small sample volumes  
Good for resource limited environments and when access to HPLC/uHPLC/HPLC-MS is limited                                                                                                               | Long incubation (24h)  
Sensitivity variable  
Range limited  
**Lack of standardisation of methodology**  
Reading imprecision (subjective)  
Unsuitable for combination therapy unless developed to incorporate tester organisms with drug-specific resistance  
Unable to distinguish native drug from active metabolite  
Itraconazole: semi-quantitative only (1)  
Generally poorer EQA performance (2)                                                                                                                                                                                                                                      |
| HPLC/uHPLC/HPLC-MS | Good precision  
Objective measurement  
Quick turnaround time (3-4 h)  
Can measure several drugs simultaneously  
May already be established in a centre and adapted for antifungal assay                                                                                                    | High equipment acquisition costs  
High maintenance costs  
High reagent costs  
Limited availability of instruments in clinical micro labs  
For some - time consuming sample preparation steps  
Best performed in batches – may increase TAT  
Requirement for skilled operator  
Need for technical support  
Possible peak interference from compounds with identical retention time (HPLC/uHPL)                                                                                                                                                                      |
| ARK Diagnostics (3) (immunoassay) | Commercially available  
Can be run on random access chemistry analysers  
Quick turn around time (5 min)                                                                                                               | High equipment acquisition costs  
Requirement for ± 120 requests/month (kit stability)  
Equipment available in biochemistry labs which lack experience on sample interpretation  
Currently only available for voriconazole                                                                                                                                                                                                                              |

2. Darville *et al.* Abstract P1693 24th ECCMID 2014

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Laboratory: How should antifungal drugs be analysed?

In-house laboratory validation and verification of the performance characteristics of the chosen method should be undertaken to include:

• Analytical specificity
• Linearity, working range and limits of detection and quantification (LOD and LOQ)
• Precision: repeatability (intra-day precision) reproducibility (intra-day precision)
• Stability of analyte on storage
• Extraction recovery (for HPLC methods)
• System suitability (for HPLC methods)
• Ongoing Internal Quality Control (IQC)
• Ongoing Internal Quality Assessment (IQA)
• **External Quality Assessment / Assurance (IQA)**

1. Shabir GA Journal of Validation Technology 2004
3. BS EN ISO 15189:2012 Medical Laboratories – Requirements for quality and competence
Laboratory: What are the external quality assurance/assessment (EQA) schemes for laboratories analysing TDM samples?

Rationale for participation in EQA schemes:

- Confirms a laboratory’s analytical method is fit for purpose
- Ensures continuing analytical competence
- Monitors ongoing accuracy
- Allows intra and inter-laboratory comparison
- Enables post-marketing vigilance for commercial test components
- Allows early recognition of potential problems
- Important to reduce potential bias in PK/PD studies
- Enhances laboratory users confidence in results

**Laboratory:** What are the external quality assurance (EQA) schemes for laboratories analysing TDM samples?

<table>
<thead>
<tr>
<th>Features to consider when selecting an EQA test</th>
<th>Ideal</th>
<th>SOR</th>
</tr>
</thead>
<tbody>
<tr>
<td>Accreditation status</td>
<td>Scheme accredited to ISO 17043 or equivalent</td>
<td>AIII</td>
</tr>
<tr>
<td>Frequency of distribution</td>
<td>Sufficient to identify performance issues in a timely manner (monthly?)</td>
<td>AIII</td>
</tr>
<tr>
<td>Range of analytes included in panel</td>
<td>itraconazole / hydroxy itraconazole</td>
<td>AIII</td>
</tr>
<tr>
<td>Range of concentrations included in panel</td>
<td>voriconazole</td>
<td>AIII</td>
</tr>
<tr>
<td>Range of concentrations included</td>
<td>posaconazole</td>
<td>AIII</td>
</tr>
<tr>
<td>Test materials</td>
<td>Commutable materials</td>
<td>AIII</td>
</tr>
<tr>
<td>Handling of performance issues</td>
<td>Mechanism in place for managing poor performance</td>
<td>AIII</td>
</tr>
<tr>
<td>Number of participants</td>
<td>Sufficient to allow significant result analysis and peer comparison</td>
<td>AIII</td>
</tr>
<tr>
<td>Management / development</td>
<td>Competent professionals</td>
<td>AIII</td>
</tr>
</tbody>
</table>

Key Questions - Laboratory: What are the external quality assurance/assessment (EQA) schemes for laboratories analysing TDM samples?

<table>
<thead>
<tr>
<th>Scheme organisation</th>
<th>INSTAND e.V.</th>
<th>KKGT</th>
<th>UKNEQAS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Accreditation status</td>
<td>Reference laboratory accredited to ISO standards</td>
<td></td>
<td>CPA accredited</td>
</tr>
<tr>
<td>Frequency of distribution</td>
<td>2 panels per year</td>
<td>2 x 2 samples per year</td>
<td>Monthly</td>
</tr>
<tr>
<td>Range of analytes included in panel</td>
<td>Itra / hydroxyitraconazole</td>
<td>Itra / hydroxyitraconazole</td>
<td>Itra / hydroxyitraconazole</td>
</tr>
<tr>
<td></td>
<td>Voriconazole</td>
<td>Voriconazole</td>
<td>Voriconazole</td>
</tr>
<tr>
<td></td>
<td>Posaconazole</td>
<td>Posaconazole</td>
<td>Posaconazole</td>
</tr>
<tr>
<td></td>
<td>Fluconazole</td>
<td>Fluconazole</td>
<td>Fluconazole</td>
</tr>
<tr>
<td>Single analyte samples suitable for</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
</tr>
<tr>
<td>bioassay</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Range of concentrations</td>
<td>Clinically relevant</td>
<td>Clinically relevant</td>
<td>Clinically relevant</td>
</tr>
<tr>
<td>Test materials</td>
<td>Commutable materials</td>
<td>Commutable materials</td>
<td>Commutable materials</td>
</tr>
<tr>
<td>Handling of performance issues</td>
<td>Certificate awarded for satisfactory performance</td>
<td>Comprehensive report provided</td>
<td>Poor performance letters / referral to oversight panel</td>
</tr>
<tr>
<td>Number of participants</td>
<td>??</td>
<td>63</td>
<td>24</td>
</tr>
</tbody>
</table>

Laboratory: What are the external quality assurance/assessment (EQA) schemes for laboratories analysing TDM samples?

EQA unmet need:

There is no interpretative EQA scheme to ensure that the correct advice is being given regarding the levels that are achieved. Or to assess any advice given on ways to attempt to rectify low or high levels.