



Meeting Title:	Subcommittee (SC) on Antifungal Susceptibility Tests	Contact:	mhackenbrack@clsi.org
Meeting Date:	Saturday, 26 January 2019	Secretary	Camille Hamula, PhD
Start Time:	8:00 AM Eastern (US) time	End Time:	2:45 PM
Location:	Legends 1 Meeting Room Renaissance World Golf Village Hotel 500 South Legacy Trail St. Augustine, Florida 32092.		
Meeting Purpose:	The purpose of this meeting is to review and discuss subcommittee business.		
Requested Attendee(s):	SC members, advisors, reviewers, all interested parties, and CLSI staff (see SC roster).		
Attendee(s):			
Gary W. Procop, MD, MS Chairholder Barbara D. Alexander, MD, MHS Vice-chairholder Camille Hamula, PhD, D(ABMM) Secretary/Advisor		Cleveland Clinic Duke University Medical Center Saskatoon Health Region/University of Saskatchewan	
Members Present:			
Philippe J. Dufresne, PhD, RMCCM Jeff Fuller, PhD, FCCM, D(ABMM) Mahmoud A. Ghannoum, PhD, FIDSA, MBA Nicole M. Holliday, BA Luis Ostrosky-Zeichner, MD, FACP, FIDSA, FSHEA, CMQ Audrey N. Schuetz, MD, MPH, D(ABMM) Nathan P. Wiederhold, PharmD Adrian M. Zelazny, PhD, D(ABMM)		Institut National de Santé Publique du Québec London Health Sciences Centre Case Western Reserve University Thermo Fisher Scientific McGovern Medical School Mayo Clinic University of Texas Health Science Center at San Antonio National Institutes of Health	
Members Excused:			
Kimberly E. Hanson, MD, MHS		University of Utah and ARUP Laboratories	
Advisors:			
David Andes, MD Elizabeth Berkow, PhD Mariana Castanheira, PhD Sharon K. Cullen, BS, RAC Tanis Dingle, PhD, D(ABMM), FCCM Scott B. Killian, BS Shawn R. Lockhart, PhD, D(ABMM) Jaques F. Meis, MD, PHD, FIDSA FRCPPath, FAAM David H. Pincus, MS, RM/SM(NRCM), SM(ASCP) Ribhi M. Shawar, PhD, D(ABMM) Dee Shortridge, PhD Paul E. Verweij, MD, FECMM Nancy L. Wengenack, PhD, D(ABMM) Sean X. Zhang, MD, PhD, D(ABMM)		University of Wisconsin-Madison Medical School Centers for Disease Control and Prevention JMI Laboratories Beckman Coulter, Inc. Microbiology Business University of Alberta Hospital Laboratory Thermo Fisher Scientific Centers for Disease Control and Prevention Canisius Wilhemina Hospital bioMérieux, Inc. FDA Center for Devices and Radiological Health JMI Laboratories Radboud University Medical Center Mayo Clinic Johns Hopkins University	
Reviewers:			
Kevin Alby, PhD, D(ABMM) Sudha Chaturvedi, PhD		University of Pennsylvania Health System New York State Department of Health	



<p>Beth P. Goldstein, PhD Stephanie L. Mitchell, PhD, D(ABMM)</p> <p>Cynthia C. Knapp, BS, MS, MT(ASCP) Sixto Leal, MD, PhD Vera Tesic, MD, MS, D(ABMM), M(ASCP) Maria M. Traczewski, BS, MT(ASCP) John D. Turnidge, MD, BS, FRACP, FASM, FRCPA Yanan (Nancy) Zhao, PhD</p>	<p>Beth Goldstein Consulting University of Pittsburgh and Children's Hospital of Pittsburgh of UPMC Thermo Fisher Scientific University of Alabama at Birmingham University of Chicago The Clinical Microbiology Institute University of Adelaide Center for Discovery and Innovation, Hackensack Meridian Health</p>
Guests (Non-Roster Attendees):	
<p>Paul Bien Alexander Bryson Hari Dwivedi Sheila Farnham Momoko Fukisaki Nilia Robles Hernandez Rita Hoffard Michael D. Huband, BS Sarah Jung Åsa Karlsson Brenda Ling Jefferey Locke Chip Oho Margaret Ordoñez Myra Townes Paula M. Snippes Vagnone, MT(ASCP) Tam Vam Matthew A. Wikler, MD, FIDSA, MBA</p>	<p>Amplex Pharma Virginia Commonwealth University bioMérieux, Inc. bioMérieux, Inc. Eiken Chemical bioMérieux, Inc. Becton Dickenson JMI Laboratories Mayo Clinic Rochester bioMérieux, Inc. Astellas Pharma Global Development Cidara Therapeutics Eiken Chemical Microbiology Institute of Colombia Duke University Medical Center MN Public Health Laboratory Harbor UCLA Medical Center IDTD Consulting</p>
Staff:	
<p>Marcy L. Hackenbrack, MCM, M(ASCP)</p>	<p>CLSI</p>



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AGENDA					
#	Start	Time	Presenter	Description	Background
Breakfast available: 7:00 - 8:00 AM (Legends 2)					
1.	8:00 AM	10 min	Dr. Procop	Opening Remarks/Introductions	N/A
2.	8:10 AM	5 min	Dr. Procop	Review/Vote on Agenda	Agenda
3.	8:15 AM	10 min	Mr. Fine	CLSI Update	N/A
4.	8:25 AM	15 min	Dr. Procop	Annual SC Update <ul style="list-style-type: none"> • Vote: January 2018 meeting summary • DOI: Updates • Rotations: 2019 Committee roster • Update: Antifungal Document status 	Presentation Roster DOI Summary January 2018 Meeting Minutes
5.	8:40 AM	60 min	S. Lockhart D. Andes N. Wiederhold	Voriconazole/ <i>Aspergillus</i> breakpoints	
6.	9:40 AM	20 min	S. Lockhart	Proposal: Standard for gradient diffusion	
	10:00 AM	15 min	Break		
7.	10:15 AM	45 min	S. Lockhart P. Dufresne	ECV WG Report Review and vote on new ECVs for M59	
8.	11:00 AM	20 min	S. Lockhart P. Dufresne	Plan for posting MIC distributions for antifungal agents with no ECVs	
9.	11:20 AM	40 min	A. Marra (Micromyx)	Rezafungin Tier 2 disk diffusion QC ranges	Request letter Presentation
	12:00 PM	60 min	Luncheon (Legends 2)		
10.	1:00 PM	40 min	M. Huband	Ibrexafungerp Tier 2 MIC QC ranges	Request letter Presentation Report
11.	1:40 PM	45 min	A. Schuetz V. Tesic	Antifungal Reporting WG report	
12.	2:25 PM	15 min	G. Procop	Other business: Outstanding Action items	Voriconazole/ <i>Candida</i> presentation
13.	2:40 PM	5 min	G. Procop	Plan for next meeting	
	2:45 PM	N/A	G. Procop	Closing Remarks and Adjournment	
	3:00 PM	N/A	Coffee and Dessert available		
	5:00 - 7:00 PM	N/A	Educational Session (Augustine C): Recent Advances in PK/PD and its Use in Setting Breakpoints		

SUMMARY MINUTES

#	Description
1.	Dr. Procop opened the meeting at 8:00 AM Eastern (US) time by welcoming the attendees and by thanking the Subcommittee (SC) participants for their continued hard work
2.	The agenda was reviewed and there were no changes. A motion to accepted agenda was made and seconded. VOTE: 8 for; 0 against; 1 absent (PASS).
3.	Mr. Glen Fine provide an update on CLSI activities. He reported that: <ul style="list-style-type: none"> • Upcoming meetings will be located at venues that are closer to airports. • Three more staff members have joined CLSI for support of microbiology committees. • M60 is now available on the free portal on the CLSI website.
4.	<p>Dr. Procop provided an update on the SC roster and activities.</p> <ul style="list-style-type: none"> • The roster of voting members remains the same except that Ms. Denise Holliday has resigned for personal reasons. The number of voting members is now at nine (9). <ul style="list-style-type: none"> – Three new advisors were added to roster: <ul style="list-style-type: none"> ○ Tanis Dingle ○ David Pincus ○ Paul Verweij – Five new reviewers were added to the roster: <ul style="list-style-type: none"> ○ Guillermo Efron-Garcia ○ Sixto Leal ○ Natasha Pettit ○ Vera Tesic ○ Nancy Zhao – A new Working Group on Antifungal Reporting was formed, and members appointed. <ul style="list-style-type: none"> ○ Audrey Schuetz (Co-Chairholder) ○ Vera Tesic (Co-Chairholder) ○ Tanis Dingle ○ Kim Hansen ○ Natasha Petit ○ Thomas Walsh ○ Nathan Wiederhold ○ Matt Wikler ○ Nancy Zhao • The January 2018 meeting summary was reviewed for vote. <ul style="list-style-type: none"> – Dr. Alexander noted several errors that needed to be corrected. It was decided that the minutes would be corrected and submitted for an electronic vote. <p>NOTE: The corrected minutes have been distributed to the voting members for review and vote by email.</p> <ul style="list-style-type: none"> • The Subcommittee’s processes were reviewed. <ul style="list-style-type: none"> – Voting members and advisors provided updates to their disclosures which were included in the meeting background material. Participants were asked to provide any potential conflicts when commenting during the meeting. – The SC voting rules were reviewed. There were eight voting members present at the meeting. Pass votes for this meeting included: <ul style="list-style-type: none"> ○ 8 - 0; 7 - 1; 6 - 3 (1 member absent) ○ 7 - 0; 6 - 2 (2 members absent or abstaining) ○ 6 - 0 (3 members absent or abstaining) • The CLSI document categories and rules for assessment and revision were reviewed. <ul style="list-style-type: none"> – Documents are categorized as Active (in the review process), Archived (available for use but not in the review process) or Withdrawn (outdated and no longer available) – Procedural documents (ie, M27, M39, M44, M51, M57) are reviewed and may be revised on a 3 to 5-year cycle. – Supplements (M59, M60, M61) can be revised annually, if needed.

- The status of Antifungal documents was reviewed.

Document #	Document Type	Edition	Publication Date	Final Due Date for Next Review	Reaffirm/ Revise/ Archive	Comments
M27 (Yeast BMD AST)	Standard	4 th	November 2017	2022	N/A	
M38 (Mould BMD AST)	Standard	3 rd	November 2017	2022	N/A	
M44 (Yeast DD AST)	Guideline	4 th	December 2018	2023	N/A	
M51 (Mould DD AST)	Guideline	1 st	10/2010	2021	Reaffirmed 2016	Upon review, determine if revision is needed; If so, a project proposal must be prepared
M57 (ECV generation)	Guideline	1 st	4/2016	2021	N/A	Can be reviewed at 3 years (2019) if revision is needed
M59 (ECV Tables)	Supplement	2 nd	January 2018	Yearly/As needed	N/A	If new ECVs are presented in Jan 2019; a revision can be started.
M60 (Yeast BMD/DD tables)	Supplement	1 st	November 2017	Yearly/As needed	N/A	If new BPs are presented in Jan 2019; a revision can be started.
M61 (Mould BMD/DD tables)	Supplement	1 st	November 2017	Yearly/As needed	N/A	If new BPs are presented in Jan 2019; a revision can be started.

- There is a call for isolates for CDC passive monitoring of azole resistance in *Aspergillus fumigatus*. A handout distributed by Dr. Elizabeth Berkow. Any number of isolates will be accepted.

5. Voriconazole and *Aspergillus fumigatus* Breakpoint

- Dr. Lockhart stated that clinical trial and outcome data is available, and the drug is in clinical use, but a breakpoint has never been set.
- Dr. Wiederhold presented clinical trial and outcome data from the Fungus Testing Laboratory, University of Texas, Health San Antonio for generating voriconazole breakpoints for *A. fumigatus*.
 - 1230 *A. fumigatus* isolates (excluding nail and ear cultures) were tested using broth microdilution (M38).
 - Identifications were confirmed by DNA sequencing.
 - The data showed voriconazole with an ECV of 1 and proposed MICs at:
 - ≤ 0.5 = S
 - 1 = I
 - ≥ 2 = R
 - Isolates with elevated azole MICs were sequenced. *Cyp51* mutations and corresponding MICs were characterized (Manuscript reference: Wiederhold et al. JCM 2016; 54). Isolates with *cyp51* mutations affecting voriconazole have MICs of 4 or higher.
 - It was noted that the bottom end of the MIC distribution for voriconazole-R isolates overlaps with high end of MIC distribution of wild type at MICs between 0.5 and 4.
 - Itraconazole data for mutants vs wild type aligns with voriconazole for the most part. Distribution of MIC data based on specific mutations is different. Dr. Fuller mentioned that for Canadian data the initial MIC distribution was comparable. Canadian sequencing data is not yet available.
 - Dr. Verweij noted that animal data suggests that the phenotype is most important.
- Dr. Andes presented pharmacokinetics/pharmacodynamics (PK/PD) data for generating voriconazole breakpoints for *A. fumigatus*.
 - Data was generated from:
 - A preclinical infection model for PK-PD
 - Clinical therapeutic drug monitoring (TDM) outcomes vs MIC distribution
 - MIC and clinical failures

- Clinical PK/PD
- Preclinical PK/PD Data
 - It was noted that triazole resistance in *Aspergillus* is important and that the animal model is important.
 - Studies in mice (Lepak et al. AAC 2013; 57:5438) showed that based on the average human PK, the susceptible MIC is predicted to be 0.25. Therapy is ineffective at higher MICs with *cyp51* mutations.
 - As you increase AUC/MIC, there is an increase in survival (Mavridou et al. AAC 2010; 54)
 - *In vitro* PKPD data (Jeans et al JID 2012: 206: 442). AUC/MIC 55, max suppression at trough/MIC ratio 1.2. *In vitro* models proposed that MICs be set at S = 0.25, I = 0.5, R = 1.0.
- Clinical TDM and MIC Distributions. Studies showed that:
 - Voriconazole PK variability intra-patient is wide. Clinical response falls off once trough falls below 1 ug/mL. Survival-trough below 2 ug/mL shows increase in mortality. Most of these patients are infected with wild-type strains with an MIC of 0.5 or less.
 - Trough was at 1-2 µg/ml and an MIC₉₀ of 0.5 µg/ml.
 - Trough to MIC of 2-4 µg/ml (total drug concentrations)
 - Trough concentration of 4 µg/ml are achievable and non-toxic
 - MIC ceiling = Trough of 4/1 = 1.0 µg/ml
- MIC and Clinical Failures
 - Higher mortality was shown in patients with infections with triazole-resistant *Aspergillus*.
- Clinical PK/PD Data
 - Retrospective, logistic regression analysis of 9 voriconazole clinical trial data showed that with an MIC range of 0.25-0.5 µg/ml, treatment was successful 65% of the time (Lestrade et al)
 - Patients with *cyp51* mutations and higher MICs have higher mortality than those infected with wild type at lower MICs. No difference seen in Heo et al. study data (this data is an exception).
 - Analysis of 9 clinical trial data sets was presented (Troke et al. AAC 2011: 55; 4782). *Aspergillus* MIC of 0.25-0.5 free AUC/MIC near 25 or total trough/MIC 2.48=MIC ceiling of 0.5.
- Conclusions
 - Elevated voriconazole MIC in *Aspergillus* matters for *in vitro* and *in vivo* models and patients
 - Susceptible BP estimates
 - MIC and outcome R ≥ 2
 - *In vitro* and *in vivo* model estimates S<0.25, R≥1
 - TDM + MIC₉₀ data R ≥ 0.25-1.0
 - Clinical PK/PD = free trough/MIC >2, R ≥ 0.5
- Discussion
 - Dr. Lockhart noted that EUCAST has set the breakpoint at 2. Dr. Verweij suggested that this may be too high as these patients often fail.
 - Dr. Alexander noted that the data was presented at least six years ago at this meeting. This data was from a comparative study with amphotericin B. At the time, there were not enough resistant isolates to appropriately set the breakpoints and there was also a lack of clinical data presented. She recalled that the MICs were presented as S ≤ 1; I = 2; R ≥ 4.
 - Dr. Ghannoum confirmed that there were only a few resistant isolates presented at that meeting.
 - Dr. Schuetz mentioned that the ECV is currently too high to apply clinically, which is how many laboratories are using it. The laboratories are under calling isolates, so it is important to set a breakpoint.
 - Dr. Procop suggested that a comment regarding the dosage levels should be included in the document along with the breakpoints.
 - Dr. Castanheira suggested that clinical trial data will never capture enough resistant isolates, so a decision should be made now rather than waiting for additional data.
 - Dr. Verweij noted that subculturing isolates to agar supplemented with azoles is helpful in making decisions to switch treatment. If the isolate does not grow, MICs are not needed. The plate is commercially available and provides an easy method for screening (EUCAST recommendation).
 - Dr. Alexander mentioned the previous discussion about whether to use SDD or I, and the “I” category was chosen. She noted that care is needed around language used for dosage levels when not using SDD. It is difficult to get trough/MIC level of 2 in patients and recommendations need to be realistic in what clinicians are expected to be able to achieve.
 - It was also suggested that a rationale document could be developed to explain the rationale for the MICs being approved.

	<ul style="list-style-type: none"> The MIC proposal made in the publication by David et al was as follows: MIC of 2 is R. MIC of 1 in intermediate, and an MIC of ≤ 0.5 is susceptible. <ul style="list-style-type: none"> It was discussed whether 1.0 should be Intermediate as it intersects ECV. Some members stated that they are concerned about MIC of 1.0. A rationale document would help explain the decisions. Ms. Cullen asked about reproducibility. Dr. Castanheira suggested that because the distributions are so different, the variability between laboratories is greater than within laboratories. Dr. Castanheira's distribution is very different from Dr. Fuller's and Dr. Wiederhold's. It was noted that there is an expected 2-fold dilution error in antifungal testing. Since IDSA does not recommend antifungal testing, testing should be emphasized in the rationale document and that IDSA should be brought into the discussion. It was agreed that the breakpoints should be set as it is unlikely that more data will become available and setting a breakpoint will encourage laboratories to perform testing. <p>A motion (Dr. Ghannoum) to accept MICs ($\mu\text{g/mL}$) for <i>Aspergillus fumigatus</i> and voriconazole ($S = \leq 0.5$; $I = 1$; $R = \geq 2$) was made and seconded (Dr. Schuetz). VOTE: 8 for; 0 against; 1 absent (PASS).</p> <ul style="list-style-type: none"> M61, <i>Performance Standards for Antifungal Susceptibility Testing of Filamentous Fungi</i> will be revised to include the new breakpoints. A simplified test and/or procedure for testing will be considered.
6.	<p>Proposal for development of a gradient diffusion standard</p> <ul style="list-style-type: none"> Dr. Lockhart proposed that since there are now two gradient diffusion strip manufacturers that a standard be developed, or the procedure be added to a current document. <ul style="list-style-type: none"> He suggested that the document could be developed as a joint project with the Subcommittee on Antimicrobial Susceptibility testing. Ms. Cullen suggested that the reference documents should be for methods where you can read the reference method and perform it. It should be considered whether most laboratories can manufacture gradient strips. Dr. Castanheira mentioned that the same issue exists with disks as manufactured disks have stabilizers and that laboratories creating their own disks may see differences in their results; however, this has not prevented development of a disk diffusion document. It was proposed that the document would focus on the methodology and it would need to be determined if the new method correlates with the reference method (broth microdilution). <p>A motion to develop a procedural document for gradient diffusion was made (Dr. Ghannoum) and seconded (Dr. Schuetz). VOTE: 7 for; 0 against; 2 absent (PASS).</p> <ul style="list-style-type: none"> Ms. Hackenbrack stated that a project proposal must be developed and approved by Consensus Council. Dr. Lockhart volunteered to develop a project proposal.
7.	<p>ECV WG Report</p> <ul style="list-style-type: none"> Dr. Dufresne presented round one ECV data for review and vote. Data was collected on the following organism/drug combinations: <ul style="list-style-type: none"> <i>Candida auris</i> (All antifungals) <i>Candida orthopsilosis</i> (All antifungals) <i>Candida kefyr</i> (All antifungals) <i>Candida dubliniensis</i> (amphotericin B, caspofungin, itraconazole, isavuconazole, flucytosine, posaconazole) <i>Candida guilliermondii</i> (amphotericin B, caspofungin, itraconazole, isavuconazole, flucytosine) <i>Candida lusitanae</i> (amphotericin B, caspofungin, voriconazole, isavuconazole, flucytosine) Seven laboratories participated in the study and raw data was compiled by the ECV WG leadership and analyzed using ECOFF Finder. <i>C. auris</i> <ul style="list-style-type: none"> Four laboratories submitted data with 99% from the CDC. There was a high level of resistance. More data from additional laboratories is needed. <i>C. orthopsilosis</i>

- Appropriate data to set ECVs was submitted for analysis of anidulafungin, micafungin, fluconazole, posaconazole, and voriconazole.
- More laboratories are needed for amphotericin B, caspofungin and isavuconazole.
- More isolates are needed for itraconazole.
- *C. kefyi*
 - Appropriate data to set ECVs was submitted for analysis of amphotericin B, anidulafungin, micafungin, fluconazole, itraconazole, and posaconazole.
 - Voriconazole and flucytosine appear truncated and additional laboratories and isolates are needed for isavuconazole.
- *C. dubliniensis*
 - Appropriate data to set ECVs was submitted for analysis of amphotericin B, itraconazole, and posaconazole.
 - Data for flucytosine, isavuconazole, and voriconazole was truncated and there was interlaboratory variation with caspofungin.
- *C. guilliermondii*
 - Appropriate data to set ECVs was submitted for analysis of amphotericin B, caspofungin, and itraconazole.
 - Additional laboratories and isolates are needed for isavuconazole and voriconazole.
- *C. lusitanae*
 - Appropriate data to set ECVs was submitted for analysis of amphotericin B and caspofungin.
 - Data from gradient diffusion for amphotericin B suggested that *C. lusitanae* may have intrinsic resistance to amphotericin B; however, no resistance was shown using broth microdilution.
 - Mechanism appears to be due to an inducible gene. It may not be seen on the first patient isolate before initiating treatment. A comment regarding testing isolates after treatment is initiated. Additional laboratories and isolates are needed for isavuconazole.

ECVs for Vote (copied from Dr. Dufresne’s presentation PDF)

Species	Antifungal	Proposed ECV	# Labs	# isolates
<i>C. dubliniensis</i>	Amphotericin B	0.5	5	461
<i>C. dubliniensis</i>	Itraconazole	0.25	5	595
<i>C. dubliniensis</i>	Posaconazole	0.125	6	722
<i>C. lusitanae</i>	Caspofungin	1	6*	580*
<i>C. lusitanae</i>	Amphotericin B	2	4	447
<i>C. guilliermondii</i>	Amphotericin B	2	4	167
<i>C. guilliermondii</i>	Caspofungin	2	5	204
<i>C. guilliermondii</i>	Itraconazole	2	4	146
<i>C. orthopsilosis</i>	Anidulafungin	2	3	145
<i>C. orthopsilosis</i>	Micafungin	1	3	145
<i>C. orthopsilosis</i>	Fluconazole	2	3	145
<i>C. orthopsilosis</i>	Voriconazole	0.125	3	145
<i>C. orthopsilosis</i>	Posaconazole	0.25	3	145
<i>C. kefyi</i>	Amphotericin B	2	4	135
<i>C. kefyi</i>	Anidulafungin	0.25	3	125
<i>C. kefyi</i>	Micafungin	0.125	4	145
<i>C. kefyi</i>	Fluconazole	1	4	129
<i>C. kefyi</i>	Itraconazole	0.5	5	111
<i>C. kefyi</i>	Posaconazole	0.5	5	154

*One lab had a mode that was low but eliminating that lab did not change the ECV

For some of the *C. orthopsilosis* and *C. kefyi* data there was one lab with >50% of the data. In each case the ECV was determined with and without adjustment of the offending lab down to 50%. IN no case was there a change in the ECV following adjustment.

- Discussion
 - o Dr. Alby noted that for VITEK MS, *C. orthopsilosis* is not claimed as there is not sufficient data for discriminating against *C. parapsilosis*. The latest 3.2 version has all three, but this version is not yet available in the US (available in Canada, US soon).
 - o For *C. lusitaniae* inducible resistance, concern was raised that the ECV may be misleading. A comment to explain the inducible resistance that may be present in the organisms should be used along with this data.

**A motion to accept the ECVs for the organisms and drugs listed in the table was made and seconded.
VOTE: 8 for; 0 against; 1 absent (PASS).**

- o Dr. Procop proposed an amendment to add a comment regarding *C. lusitaniae*. Dr. Schuetz proposed omitting the *C. lusitaniae* ECV and comments for the working group on reporting to address. All voting members were in favor of Dr. Schuetz's proposal

• ECV Data Round 2 Update

- New *Candida* spp. for data collection were selected.
 - o Rare members of *Candida haemulonii/auris* complex
 - o *Candida duobushaemulonii*
 - o *C. parapsilosis* (*C. metapsilosis*)
 - o *Lodderomyces elongisporus*
 - o *Candida rugosa*
 - o *Candida pararugosa*
 - o *Candida bracarensis*
 - o *Candida nivariensis*
- Data for all antifungals was requested. For many antifungals, there are close to 20 isolates but a minimum of 100 isolates is needed. Rare species were selected based on their prevalence and if they were included in a species complex.
- Nine laboratories have indicated that they could or have already provided data.
- To date, less than 100 isolates of each species have been submitted.
- CLSI reference panels can be purchased from ThermoFisher if your laboratory does not perform the CLSI method but has isolates. Laboratory should look for more funding for ThermoFisher panels.
- The ECV WG requested that data be submitted to Dr. Dufresne or Dr. Lockhart by the spring 2019.
- Plans for collecting and analyzing *C. auris* isolates were discussed.
 - The CDC currently has MICs for more than 750 isolates with an additional 40 isolates to be tested.
 - MICs for approximately 50 isolates are available from the Patel Chest institute.
 - Multimodal distributions have been generated to date.
 - Additional data may provide more definitive ECVs.
- A list of proposed species for Round 3 were presented. Other suggestions should be sent to Dr. Dufresne or Dr. Lockhart.
 - *Geotrichum candidum*
 - *Saprochaete clavata*
 - *Saprochaete capitata*
 - *Trichosporon (inkin, asahii)*
 - *Rhodotorula (mucilaginoso, minuta)*
 - *Saccharomyces cerevisiae*

8. Plan for posting MIC distributions for antifungal agents with no ECVs

- Dr. Dufresne restated the participants of the proposal for posting MIC distribution for those antifungal agents for which there are no ECVs. Gaps in ECVs are due to:
 - Insufficient number of laboratories or isolates
 - Truncated distributions
 - Abnormal multimodal MIC distributions
- Current criteria for setting ECVs:
 - Minimum 20 isolates
 - 3 labs (may be bypassed for very rare species)

- Testing using methods in M27 or M38
- ID by MALDI-TOF MS or sequencing
- QC strain data must be provided and within acceptable range
- The proposal that was accepted during the January 2018 meeting was for the following antifungal MIC distributions to be published on the CLSI Website
 - For rare species (with not enough data or laboratories)
 - For truncated distributions
 - Abnormal, multi-modal distributions **would not** to be published
- The accepted criteria for publishing MIC distributions were restated.

Criteria	ECV (M57)	Publication of MIC distribution with no ECV (proposal)
Minimal MIC data points (1 clinical strain/patient)	Min. 100	Min. 20.
Number of submitting labs	Min 3 labs (weighed if needed)	Min 3 labs (weighed if needed) No minimum for rare species
Methodology	CLSI M27 or M38	CLSI M27 or M38 (reading time and inoculum may need to be disclosed)
Species identification method	Molecular confirmed by MALDI-TOF or sequencing (ECV WG may be more specific for some species which are more difficult to ID)	Molecular confirmed by MALDI-TOF or sequencing (ECV WG may be more specific for some species which are more difficult to ID)
QC strains data	Must be provided and within accepted range	Must be provided and within accepted range
Truncated dataset	Not accepted	Accepted if they cover recommended CLSI M27 and M38 concentration ranges for a given antifungal agent.
Dataset that are not within 1-2 dilution of pooled dataset	Reviewed as potential outliers, rejected if it is the case.	Reviewed as potential outliers, rejected if it is the case. Not applicable in some case where the number of labs is too small
Abnormal or multimodal MIC/MEC lognormal distribution	Rejected	Rejected But a note would be included to MIC distribution listing

- Discussion
 - Ms. Cullen suggested that it needs to be determined if outliers are “real” and not due to error on the part of a laboratory. Differences may be due to organism differences or reproducibility issues with a testing methodology. It was suggested that an organism set be created to help laboratories investigate and verify their methods if their data are flagged as out of range. The CDC is working on an organism panel; however, the majority are bacteria, but fungi are continuously being added. CDC will distribute the set internationally. Dr. Jean Patel is coordinating on bacterial side.
 - Dr. Lockhart suggested that reproducibility data by laboratory could be published. Dr. Castanheira suggested that the testing should be repeated for the outliers before proceeding to publication.
- Originally studied organisms with MICs that could be published now include:
 - *C. albicans*: 5FC
 - *C. glabrata*: 5FC, ITRA
 - *C. krusei*: 5FC
 - *C. parapsilosis*: 5FC, ITRA
 - *C. tropicalis*: 5FC
- Round 1 organism MICs that could be published include (red/underlined = truncated distribution):
 - *C. orthopsilosis*: 5FC, AMB , CAS , ITRA
 - *C. kefyri*: 5FC, VORI
 - *C. dubliniensis*: 5FC, VORI
 - *C. guilliermondii*: 5FC, VORI
 - *C. lusitaniae*: 5FC, VORI
- Round 2 organism MICs that could be published include:
 - *C. bracarensis*: ALL (except 5FC)

- *C. nivariensis*: ALL (except 5FC)
- *C. duobushaemulonii*: ALL (except 5FC)
- *C. haemulonii*: ALL (except 5FC)
- *C. metapsilosis*: 5FC, AMB, ITRA
- *L. elongisporus*: ALL (except 5FC)
- *C. pararugosa*: ALL (except 5FC)
- *C. rugosa*: ALL (except 5FC)
- A potential format for presenting the MIC distributions was discussed.
 - A table created and organized by antifungal agent or by microorganism, would include the number of laboratories with data, the number of MICs, and columns stating why no ECV is available.
 - A second option would be to present the data as a histogram.
- The location where the data will be posted was discussed.
 - Data public or only accessible to those with a CLSI subscription/membership?
 - CLSI exchange, new dedicated web page, current “Antifungal educational web page”?
 - <https://clsi.org/meetings/sub-antifungal/>
 - Referenced in our other CLSI documents (M57/59, M27/60, M38/61)?
 - Ms. Hackenbrack stated:
 - If the information is to be available to anyone, regardless of their association with CLSI, it could be presented on the Antifungal Subcommittee page on the CLSI Website which is open to anyone.
 - If the information is to be restricted to the Antifungal Subcommittee roster participants, it could be posted on CLSI Exchange. Only those listed on the roster have access to information posted on exchange.
 - In either case, the site could be referenced in CLSI documents; however, data on Exchange would not be available to everyone using the documents.
 - Ms. Hackenbrack requested that the SC submit a proposal for what will be posted and to whom it will be available so that the distributions can be appropriately managed.

9. **Rezafungin Tier 2 Study for Disk Diffusion QC Ranges**

- Drug is currently in Phase 3 for treating candidemia
- Study data for setting disk diffusion QC ranges for rezafungin (novel echinocandin) was presented.
 - Drug with potent *in vitro* activity against *Candida* spp.
 - 5 µg disk shown to be optimal correlation with broth microdilution and was able to differentiate *fks* mutants from other species.
 - Tested in nine laboratories using CLSI M44/M60 methods, three lots of media, two lots of disks from one manufacturer and with caspofungin as the control agent.
 - Isolates tested:
 - *Candida parapsilosis* ATCC 22019
 - *C. krusei* ATCC 6258
 - *C. albicans* ATCC 90028
 - *C. tropicalis* ATCC 750
 - Based on the study data, the sponsor requested the following QC ranges be approved.

Organism	Proposed QC Range (mm)
<i>C. parapsilosis</i> ATCC 22019	9 - 16
<i>C. krusei</i> ATCC 6258	14 - 20
<i>C. albicans</i> ATCC 90028	13 - 20
<i>C. tropicalis</i> ATCC 750	14 - 20

- Discussion
 - Ms. Cullen provided a summary of the presented QC information in the same format as used by the Antimicrobial Susceptibility Testing Subcommittee (AST). She recommended that the summary include:
 - Solvent and diluent information
 - Information based on M23 requirements
 - Calculate with traditional methods and Rangefinder method

- For disk: Gavan statistic based on median with expansion if less than 95% of result is included
- For MICs: Mode \pm 1 dilution, expand to 4-dilution range if shoulder is >60% or <95% of result included
- Laboratory data can be excluded if there is a statistical outlier for 2-3 parameters (mean, median, mode). Don't exclude if outlier for only one parameter.

– **Summary for rezafungin**

Drug: Rezafungin	Abbreviation: ?	Previous ID: ?
Solvent: ?	Diluent: ?	Preparation: ?
Route of administration:	Class: novel echinocandin	Subclass:
Study Report by: MicroMyx	Pharma Co: Cidara Therapeutics	Control Drug: caspofungin

Footnotes:	Add footnote “QC ranges for Rezafungin was established using data from only one disk manufacturer. Disks from other manufacturers were not available at the time of testing.”
Discussion	Similar <i>in vitro</i> activity to anidulafungin against <i>Candida</i> spp. Disk masses of 5, 10, 15, 20, 25 μ g evaluated. 5 μ g differentiated wild-type vs <i>fks</i> mutants. Linear correlation coefficient R = -0.9074. Control drug caspofungin >95% in range except for <i>C. tropicalis</i> ATCC 750 with only 88.1% in range. All were out high with Media B and C. Do we need to reevaluate the QC range for this strain (Tier 3)?

QC Strain	Range	% In	Median	mm	Comments
<i>C. parapsilosis</i> ATCC 22019	9-16	94.9%	13	8	Excluding QC out of range control Lab 7: was it statistical outlier for 1 or 2 parameters? - don't remove if 1 parameter 99.4% excluding out of range control and Lab 7. Gavan 99.1% excluding Lab 7.
<i>C. krusei</i> ATCC 6258	14-20	99.1%	17	7	
<i>C. albicans</i> ATCC 90028	13-20	100%	17	8	98.5% in range before excluding out of range QC
<i>C. tropicalis</i> ATCC 750	14-20	99.2%	17	7	Excluding QC out of range control Slide with Lab 3 & 5 removed - were these outliers for 2 parameters?

A motion to accept the QC ranges presented for rezafungin was made (Dr. Wiederhold) and seconded (Dr. Zelazny). VOTE: 7 for; 0 against; 2 absent. (PASS).

10. **Ibrexafungerp Tier 2 Study for Broth Microdilution QC Ranges**

- Study data for setting MIC QC ranges for ibrexafungerp were presented by Mr. Huband.
 - CLSI M23 (2018) Tier-2 criteria were followed.
 - Eight laboratories participated (\geq 7 laboratories required)
 - Three lots of RPMI 1640 medium base obtained from at least 2 (3) different manufacturers were used.
 - Strains tested
 - *Candida parapsilosis* ATCC 22019 (24 and 48h)
 - *Candida krusei* ATCC 6258 (24 and 48h)
 - Based on the study data, the sponsor requested the following QC ranges be approved.

Reference strain	Proposed CLSI QC range (μ g/mL)	
	IBX 24 hour MIC	IBX 48 hour MIC
<i>Candida parapsilosis</i> ATCC 22019	0.06 - 0.25 (3; 99.0%)	No range proposed
<i>Candida parapsilosis</i> ATCC 22019	N/A	0.12 - 0.5 (3; 100.0%)
<i>Candida krusei</i>	0.25 - 1	0.25 - 1

ATCC 6258	(3; 100.0%)	(3; 100.0%)
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– Summary for Ibrexafungerp

Drug: Ibrexafungerp	Abbreviation: ?	Previous ID: SCY078
Solvent: ?	Diluent: ?	Preparation: ?
Route of administration: ?	Class: ?	Subclass: ?
Study Report by: JMI	Pharma Co: ?	Control Drug: anidulafungin

Footnotes:	
Discussion	Colony count ave 1.6×10^3 CFU/ml. Is this low or normal for these strains?

QC Strain	Range	% In	Mode	dil	Comments
<i>C. parapsilosis</i> ATCC 22019 - 24 hr	0.06-0.25	99.0%	1	3	Lab E excluded as outlier for mean, median, mode (≤ 0.15) for control drug.
<i>C. parapsilosis</i> ATCC 22019 - 48 hr	No range	NA	NA	NA	Alternative 0.12-0.5 but only 6 labs after excluding Lab E (see above) and Lab C as outlier for mean, median, and mode (4)
<i>C. krusei</i> ATCC 6258 - 24 hr	0.25-1	100%	0.5	3	
<i>C. krusei</i> ATCC 6258 - 48 hr	0.25-1	100%	0.5	3	

- Discussion
 - The *C. parapsilosis* 48h value had QC outliers for Lab E. Lab C was a complete outlier and must be removed. If both are removed, there will not be 7 labs as required by M23 criteria. Lab E data has 30 values. Discuss either no proposed range or include Anidulafungin QC data from Lab E that was out to propose 0.12-0.5 range. QC outliers from Lab E likely due to reading error.
 - It was agreed that a 48 hr range is needed for *C. parapsilosis* ATCC 22019.
 - Options for the approval of the ranges presented were:
 - o Wait for the sponsor to submit additional information and approve all at the same time
 - o Approve the available ranges and provide a reason for why there is no 48 hr. range for *C. parapsilosis*
 - o Approve the available ranges and request additional data for a 48 hr. range for *C. parapsilosis*
 - It was agreed that the available ranges would be approved, and the sponsor will be asked for additional data to approve a 48 hr. range for *C. parapsilosis*.

A motion to accept the available QC ranges (24 and 48 hr for *C. krusei* ATCC 6258 and 24 hr. for *C. parapsilosis*) with a request for additional data from another laboratory for *C. parapsilosis* at 48 hr. was made and seconded. NOTE: The new data will be presented separately and combined with the original data. VOTE: 8 for; 0 against; 1 absent (PASS).

11. Antifungal Reporting WG Report
 Roster: Audrey Schuetz, Vera Tesic (Co-Chairholders); Tanis Dingle (Recording Secretary); Kimberly Hanson, Stephanie Mitchell, Natasha Pettit, Thomas Walsh, Nathan Wiederhold, Nancy Zhao (Members).

- The charge of the WG was presented. The WG has begun preliminary discussions and expect formal work to begin in 2019.
 - To develop guidelines for reporting of certain antifungal agents from specific body sites (and, conversely, those body sites from which certain antifungals would not be appropriate to report)
 - To explore options for restriction of reporting of certain antifungal agents considered intrinsically resistant to certain fungi
- Restricting echinocandin and voriconazole reporting for *Candida* isolates from urine

	<ul style="list-style-type: none"> - The IDSA 2016 Clinical Practice Guidelines for the Management of Candidiasis recommends that echinocandins and voriconazole not be reported for urine candidiasis as the agents do not achieve therapeutic concentrations in the urine. - Currently, CLSI does not provide guidance on restrictive reporting by body site for antifungal agents. <ul style="list-style-type: none"> o Clinical laboratories do discuss testing and reporting options with providers and the antimicrobial stewardship team. o It was proposed that CLSI recommend restricting reporting of voriconazole and/or echinocandins from urinary sources o For <i>Candida</i> urinary sources, it was suggested to test and report on request only. If requested, a comment would be included in the report (eg, Echinocandins are not considered adequate for treatment of urinary candidiasis.” OR “Echinocandins achieve limited drug concentrations in the urinary tract.”) o Exceptions have been reported such as limited success with echinocandin treatment of symptomatic candiduria (with source control, when possible) for non-<i>C. albicans</i> infections of the higher urinary tract. - Points to consider for body site restriction <ul style="list-style-type: none"> o Restriction of certain antifungals o Restriction based on body site (eg, urine) o Reporting (Never report or report upon request but add a “qualifier comment”) • Intrinsic resistance reporting for antifungal agents <ul style="list-style-type: none"> - There are several current examples of intrinsic resistance to antifungal agents <ul style="list-style-type: none"> o <i>C. krusei</i> and fluconazole o <i>Aspergillus terreus</i> and amphotericin B o <i>Aspergillus fumigatus</i> and fluconazole - Others to consider <ul style="list-style-type: none"> o <i>C. lusitanae</i> resistance to amphotericin B (intrinsic and acquired) o <i>Cryptococcus</i>: echinocandins o <i>Rhodotorula</i>: azoles and echinocandins o <i>Trichosporon</i>: echinocandins o Mucorales: voriconazole and echinocandins o <i>Scedosporium apiospermum/P. boydii</i>: echinocandins - It was noted that there are comments regarding intrinsic resistance in M27 (subchapter 3.4.3) and M60 (Tables 1 and 5 footnotes). It was suggested that a reporting comment may be needed (eg, This antifungal agent is not considered adequate for treatment) or a recommendation that the laboratory report the isolates as intrinsically resistant. - General points to consider for intrinsically resistant isolates <ul style="list-style-type: none"> o Always report as R? (What if a lab does not usually test the organism/drug combination) o If tests as susceptible, change to R? o What if a breakpoint does not exist, and only ECVs are available. How should this be reported? - Dr. Alexander suggested that a guidance document on intrinsic resistance could be published.
12.	<p><u>Other business</u></p> <ul style="list-style-type: none"> • <i>C. glabrata</i> and Voriconazole data review <ul style="list-style-type: none"> - Dr. Alexander and Dr. Fuller reviewed the original data from 2006 on clinical MIC outcomes for all <i>Candida</i> spp. but not for individual species. - No new data is available to justify changing what is already published. - It was noted that there were issues with the way the studies were performed. - It was noted that it might be possible to look at stored isolates from clinical trials. - It was concluded that there are insufficient data to move forward. - A summary of recent literature with data on voriconazole, <i>C. glabrata</i> and outcome <ul style="list-style-type: none"> o Sanchis, Castanheira, et al. Int J Antimicrob Agents.2016 <ul style="list-style-type: none"> ▪ Study challenged 27 <i>C. glabrata</i> against vori dosed 40 mg/kg in neutropenic mouse model of disseminated infection ▪ fungal burden reduction in kidneys used as marker for efficacy ▪ Voriconazole reduced burden for MICs < ECV of 0.25 ▪ Efficacy at 0.25 was variable ▪ Voriconazole was not effective for MICs >ECV of 0.25

- Rodriguez et al. PLOS.2017. *Candida* BSI and Outcomes: Authors do not link MIC data with clinical outcome, although both are presented independently
- Hirano et al. Infect Drug Resist. 2018;11:821: Contains a lot of outcome, species, and breakthrough infection data but the authors do not correlate MIC and outcome
- Patel, JCM, 2018
 - Nice current review and summary of the existing data
 - authors did not identify new evidence to support/refute correlation of Vori MIC with outcome in *C. glabrata*
 - recapitulated that Vori has a limited role in IC based on:
 - paucity of clinical outcome data linked to MIC or PKPD
 - TDM needed with Voriconazole and there are no *C. glabrata*-specific data to establish a therapeutic window
- Dr. Lockhart mentioned that often the studies use first isolates from the patient, when in fact the resistance is inducible. The way that the studies are done needs to be changed in order to resolve this issue.
- Dr. Castanheira proposed doing a project using isolates from after therapy is initiated.
- Item is completed, and will be tabled for now and removed from action item list
- *Aspergillus nidulans* data request will be tabled
- Fungal nomenclature
 - Dr. Lockhart stated that clarification is needed on fungal nomenclature. Mass spectrometry and sequencing databases may use teleomorph names. He proposed that CLSI documents should include these names in addition to anamorph name or provide clarification. He noted that this is particularly important for yeasts.
 - Dr. Alexander noted that a mycology study group is planning a discussion of changes in nomenclature as well as what an organism is also known as. CLSI may wish to form a WG to study the issue.
 - Dr. Ostrosky-Zeichner questioned whether laboratories can keep pace with nomenclature changes.
 - Dr. Fuller mentioned that clinical laboratories can modify this in their LIS, and CLSI might need a table that correlates teleomorphic and anamorphic names so that clinical laboratories can modify their reports
 - Dr. Schuetz mentioned that CLSI expert panel proposed providing guidance on taxonomy (for bacteria also) and it was voted down by Consensus council. Consensus council's view is that there are already resources available.
 - Dr. Shawar suggested perhaps this can be a function of the Outreach work group.
 - Dr. Procop stated that it is in our purview to supply clarifying information in any of our current documents.
 - Dr. Alexander mentioned that it is worth having a supplement for nomenclature that is updated annually. She proposed assembling a working group to deal with this issue and recommend which name to report.
 - Dr. Shawar note that there is a streamlined process to address this at FDA providing manufactures leeway to update their systems without having to resubmit.
 - It was questioned if a nomenclature table with the current names and what it is also known as could be created and included in antifungal documents. A WG on nomenclature could be formed to study the problem and decide how to address it.
- 21st Century Cures
 - Dr. Lockhart noted that you can go online and match CLSI breakpoints to FDA breakpoints. Most of the time FDA refers to CLSI. For itraconazole and flucytosine, the FDA Website mentions M27-S3, whereas M27-S4 specifically states that itraconazole and flucytosine breakpoints should not be used and are incorrect.
 - Dr. Shawar noted that the Website is updated quarterly and Ms. Hackenbrack noted that M60 is now referenced ([FDA STIC Antifungal](#)).
 - Dr. Procop and Dr. Alexander will work with Dr. Lockhart to draft letter to the FDA regarding the error.
 - Ms. Cullen suggested that a rationale document might provide insight.

	<ul style="list-style-type: none"> • Dr. Fuller stated that there are typographical errors in M61 that need to be corrected. <ul style="list-style-type: none"> – Ms. Hackenbrack suggested that corrections could be made when the supplement is revised to include the new <i>Aspergillus</i> breakpoints. – Dr. Fuller also noted that historical records for M61 were not well-maintained and suggested to include QC tables similar to the ones Ms. Cullen has developed for the minutes going forward. • Outreach WG Report <ul style="list-style-type: none"> – Dr. Castanheira provided a report on the activities of the AST Outreach WG. She serves as the antifungal liaison to the Outreach WG. – The Outreach WG is responsible for providing: <ul style="list-style-type: none"> ○ Educational workshop organization ○ Newsletter ○ Guidance for new CLSI participants ○ Educational documents – Dr. Castanheira provided examples of educational articles published in the most recent AST newsletter. <ul style="list-style-type: none"> ○ <i>Candida auris</i> ○ When to perform antifungal testing on <i>Candida</i> spp. isolated from patient specimens – It was proposed that the Antifungal subcommittee work with the Outreach WG to: <ul style="list-style-type: none"> ○ Communicate the activities of the Antifungal Subcommittee to the CLSI community ○ Discuss issues that are important to the clinical microbiology/mycology community – Ideas for mycology articles for inclusion in future newsletters were requested. Some examples: <ul style="list-style-type: none"> ○ Voriconazole breakpoints ○ When to test filamentous fungi ○ Antifungal pipeline – Dr. Alexander recommended that topics be assigned to SC members. – Dr. Procop also suggested that the nomenclature topic could also be added.
13.	<p>Plan for next meeting</p> <ul style="list-style-type: none"> • The next meeting of the Subcommittee will be held by Web conference in late spring or early summer. I poll will be distributed.
14.	Dr. Procop thanked the participants for their attention and continued hard work. The meeting was adjourned at 2:45 PM Eastern(US) time.

Upcoming Meetings of the Subcommittee on Antifungal Susceptibility Tests:

16 - 18 June 2019 at the Westin Galleria, Dallas, Texas, USA (potential) or Web conference (if needed)

26 - 28 January 2020 at the Tempe Mission Palms Hotel, Tempe, Arizona, USA (registration and hotel block will open in October)

14 - 16 June 2020 at the Hyatt Regency Baltimore Inner Harbor, Baltimore, Maryland, USA or Web conference (if needed)

ACTION ITEMS			
#	Description	Responsible	Status
1.	Revise M59 (new ECVs), M60 (new QC ranges), and M61 (new breakpoints and correct errors).	M59 - ECV WG M60 and M61 - WG to be formed	In progress
2.	Distribute the January 2019 Summary minutes for review and vote.	Ms. Hackenbrack	Completed Summary approved
3.	Submit isolates and/or data for the listed Round 2 ECVs to Dr. Lockhart and Dr. Dufresne	All	In progress
4.	Submit suggestions for Round 3 ECVs to Dr. Lockhart and Dr. Dufresne	All	In progress
5.	Submit a formal proposal for what MIC distributions will be posted and to whom it will be available so that the distributions can be appropriately managed.	ECV WG	
6.	Develop guidelines for reporting of certain antifungal agents from specific body sites (and, conversely, those body sites from which certain antifungals would not be appropriate to report)	Reporting WG	
7.	Explore options for restriction of reporting of certain antifungal agents considered intrinsically resistant to certain fungi	Reporting WG	
8.	Draft a project proposal for a guideline on gradient diffusion	S. Lockhart	
9.	Develop project proposal for a document on changing nomenclature	TBD	
10.	Draft a letter to the FDA regarding any errors on the FDA website	G. Procop B. Alexander S. Lockhart	

Respectfully submitted,
 Marcy L. Hackenbrack, MCM, M(ASCP)
 Camille Hamula, PhD, D(ABMM)