LABQUALITY

External Quality Assessment Scheme

General Bacteriology 1
Round 2, 2017

Please find enclosed the results of the round. The four specimens of this distribution were sent to 47 laboratories, altogether from 14 countries. Additionally, in the General Bacteriology 2 scheme, including specimens 001 and 002 solely, participated 30 laboratories out of 9 countries.

The specimens were as follows:

Specimen 001: *Streptococcus dysgalactiae* ssp. *equisimilis* (group G) ATCC® 12394™

Specimen 002: *Pseudomonas aeruginosa* CL 90-7334

Specimen 003: *Candida glabrata* N000005

Specimen 004: *Clostridium tertium* ATCC® 14573™

Results
The results of the round are presented in summary tables.
- Final report to the clinician. Enclosed also a summary table of the results reported by laboratories participating in the General Bacteriology 2 scheme. The grey areas show the laboratories´ own results. Please check that the client code on the printouts is correct.
- Susceptibility testing results by disk diffusion method of specimen 001 are shown in numerical summary. Laboratory specific histograms are drawn for each antimicrobial agent if the laboratory's result is included in a group of at least three results. By "group" is indicated results which are obtained and interpreted according to the same standard (e.g. EUCAST, CLSI etc.). The MIC-results are shown in Annex 1.

For laboratories ordering paper prints: The laboratory-specific numerical summaries, histograms and report letter of this round are also available on the Labquality homepage www.labquality.fi. Please choose Login to LabScala on the top right-hand corner and fill in your laboratory client code/personal user name and password. Next choose *View Reports*.

Comments
Specimen 001
Background information: Peritonsillar abscess developed after tonsillitis treated with erythromycin. Ongoing clindamycin treatment.
Finding: *Streptococcus dysgalactiae* ssp. *equisimilis,* Group G, as a significant pathogen.

Patient and specimen
Group G streptococci (*S. dysgalactiae* ssp. *equisimilis)* can cause tonsillitis and peritonsillar abscess.

Culture of the specimen
When tonsillitis is suspected, specimen should be cultured using agar suitable for streptococci, possibly even selective agar. When complicat-

2017-08-04

FINAL REPORT

Product no: 5080
LQ760117021-024/US
UN3373

Subcontracting: sample pre-testing

Items dispatched:	2017-05-09
Closing date:	2017-06-05
Expected results:	2017-06-07
Final report:	2017-08-04

The report includes
- the expected results
- comments on the results by the scheme expert
- laboratory specific tables

Request for correction
Typing errors on laboratory's result forms are on laboratory's responsibility. Labquality accepts responsibility only for the result processing.
Requests for correction must be notified in writing within one month of receiving the results.

Next round
The next General Bacteriology 1 EQA round (3, 2017) will be carried out in September 2017.

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Quality Management LQP ISO 9001

ed and under treatment, specimen should be cultured on both blood agar and chocolate agar plates, and possibly (if the specimen is obtained from abscess by puncture) also anaerobically incubated plates.

Identification

Only one pathogen was isolated from the specimen. Colony morphology was typical for beta-hemolytic *Streptococcus*. MALDI TOF based methods, as well as biochemical test such as Vitek GP ID Card identified this beta-hemolytic bacteria as *Streptococcus dysgalactiae*. These methods poorly differentiate two subspecies *equisimilis* and *dysgalactiae*, latter however being only rarely isolated from human specimens. The presence of Lancefield group antigen G was detected by commercial Lancefield antigen grouping sera.

Table 1. Summary of results, specimen 001.

Finding	All participants	Reference group*
No. of returned results / No. of participants	75/77 (97 %)	46/47 (98 %)
Streptococcus dysg.* ssp. *equisimilis	21/75 /28 %)	**14/46 (30 %)**
***Streptococcus* sp. , beta-hem., group G**	39/75 (52 %)	**23/46 (50 %)**
Significant pathogen	52/60 (87 %)	33/37 (89 %)
Possible pathogen	7/60 (12 %)	3/37 (8 %)
Streptococcus dysg. ssp. *dysgalactiae*	10/75 (13 %)	8/46 (17 %)
False identifications[1]	5/75 (7 %)	1/46 (2 %)

*****Participants of the General Bacteriology 1 scheme

[1] *Streptococcus* sp., beta-hemolytic, group C (1), Group A (1), *S. pyogenes* (1), *S. agalactiae* (2).

Comments on susceptibility test results

This group G streptococcus strain had no acquired resistance to any of the relevant drugs.

With a very few exceptions, laboratories correctly reported the strain as susceptible. In addition, albeit of misleading background information, majority (62/67) of the laboratories reported the strain also as MLS_B-negative. The few false erythromycin-R interpretations and (consecutive) false positive MLS_B interpretations are signs of quality problem in susceptibility testing in some of the laboratories. These laboratories should carefully review their procedure to correct it.

In fact, the original idea was to include an MLS_B-positive strain in this round. Due to some misunderstandings between the supplier of the specimens and us, this could not be realized.

Table 2. The MIC-results reported of *Str. dysgalactiae* ssp. *equisimilis* ATCC® 12394™, by two Finnish reference laboratories. Both laboratories implement the EUCAST standard.

Antimicrobial agent	**Ref. laboratory 1** **MIC (mg/L)**	**SIR**	**Ref. laboratory 2** **MIC (mg/L)**	**SIR**
Clindamycin	0,25	S	0,19	S
Erythromycin	0,125	S	0,094	S
Penicillin	0,032	S	0,016	S
Other results	**Report**			
Inducible MLS_B resistance	No		No	

Specimen 002

Background information: Keratitis. Corneal sample.

Findings: *Pseudomonas aeruginosa* as a significant pathogen.

Patient and specimen

P. aeruginosa can cause a difficult keratitis. It is not a part of normal microbiota of cornea or conjunctiva. Hence, it should be considered as a significant pathogen when isolated from specimens obtained from eye.

Culture of the specimen

Specimen should be cultured on both blood agar and chocolate agar plates.

Identification
Only one pathogen was growing well on both plates. Gram staining revealed straight and rather long and narrow gram negative rods. Oxidase reaction was positive. Isolate produced green pigment and had an odor typical for *P. aeruginosa.*
MALDI TOF based methods, as well as biochemical tests such as Vitek-2 GN ID Card can be used in identifying *P. aeruginosa*.

Table 3. Summary of results, specimen 002.

Finding	Numbers of reported results within group	
	All participants	Reference group*
No. of returned results / No. of participants	75/77 (97 %)	46/47 (98 %)
Pseudomonas aeruginosa	**73/75 (97 %)**	**45/46 (98 %)**
Significant pathogen	68/73 (93 %)	43/45 (96 %)
Possible pathogen	4/73 (5 %)	1/45 (2 %)
Pseudomonas sp.	1/75 (1 %)	1/46 (2 %)
False identifications[1]	1/75 (1 %)	0/46

*Participants of the General Bacteriology 1 scheme.
[1] *Cedecea davisae* (1)

Specimen 003
Background information: Hospital-acquired infection (sepsis). As sample is the tip of the venous cannula.
Finding: *Candida glabrata* as a significant pathogen.

Patient and specimen
Candida species are common cause of bloodstream infections associated with cannulas.

Culture of the specimen
Specimen should be cultured on both blood agar and chocolate agar plates.

Identification
Only one pathogen was growing well on both plates. Colony morphology was typical for yeast. Chromogenic agars can be used in identification. Also MALDI TOF based methods, as well as biochemical tests such as Vitek-2 YST ID Card can be used in identifying *C. glabrata*.

Table 4. Summary of results, specimen 003.

Finding	Number of reported results
No. of returned results / No. of participants	46/47 (98 %)
Candida glabrata	**41/46 (89 %)**
Significant pathogen	37/41 (90 %)
Possible pathogen	3/41 (7 %)
Yeast/*Candida* sp., other than *C. albicans*	3/46 (7 %)
Candida sp.	1/46 (2 %)
False identifications[1]	1/46 (2 %)
Additional findings[2]	3/46 (7 %)

[1]*Candida tropicalis* (1).
[2]*Staphylococcus capitis* (1)*, S. epidermidis* (1)*, S.hominis* (1).

Specimen 004
Background information: Peritonitis in a patient with neutropenia. Ascitic fluid sample.
Findings: *Clostridium tertium* as a significant pathogen.

Patient and specimen
C. tertium causes infections mainly in immunocompromised host. Many bacterial species can cause peritonitis in neutropenic patient.

Culture of the specimen
According the specimen details, this kind of specimen should be cultured using both aerobically and anaerobically incubated plates.

Identification

Only one pathogen was isolated from the specimen. Gram stain showed large, poorly staining gram positive rods, with occasional terminal spores. Combined with aerotolerancy this suggested *C. tertium*.

MALDI TOF based methods, as well as biochemical test such as RapID ANA II test kit and Vitek ANC Card can be used in identifying *C. tertium*.

Table 5. Summary of results, specimen 004.

Finding	Number of reported results
No. of returned results / No. of participants	46/47 (98 %)
Clostridium tertium	**35/46 (76 %)**
Significant pathogen	32/35 (91 %)
Possible pathogen	2/35 (6 %)
Clostridium sp.	4/46 (9 %)
Anaerobe gram positive rod	3/46 (7 %)
False identifications[1]	4/46 (9 %)
Additional findings[2]	4/46 (9 %)

[1] *Clostridium ramosum* (1), *Streptococcus mutans* (1), *Sphingomonas paucimobilis* (1), anaerobe gram negative rod (1).

[2] *Edwardsiella ictaluri* (1), *Ewingella americana* (1), *Tatumella ptyseos* (1), *Leuconostoc mesenteroides* (1).

Preanalytical case, specimen 004

Information given

Peritonitis in a patient with neutropenia. Ascitic fluid sample. The sample arrives to the laboratory in a tightly sealed, half-filled tissue sample jar with a screw cap, on the sample collection day.

Comments on results

There was preanalytical error with the specimen. Namely, wrong container had been used. When this type of container is only half filled, the specimen is in contact with air. Consequently, strictly anaerobic bacteria may not survive.

If second sample is not possible to get and specimen is analyzed, preanalytical error should be commented in the result.

67% of the laboratories that took part in this preanalytical section commented that there was preanalytical error present. Many commented that wrong container made anaerobic culture unreliable.

Scoring

General rules

Scoring is implemented for each finding when ≥60% of the laboratories participating in the General Bacteriology **1** scheme report a correct/expected result. These laboratories are considered as a reference group also for the laboratories participating in the General Bacteriology 2 scheme. **The scoring range/finding is 0-5 points.**

The scoring comprises the following elements:

- species identification, a maximum of 4 points is given
- the interpretation of the significance of the finding, a maximum of 1 point is given
- in case of insufficient species identification; an additional score (maximum 1 point) might be given to participants that would have referred the isolate for further identification

The following general rules are followed regarding the scoring of the findings:

- 4p. (maximum score) is reached by reporting the expected correct result, or, by reporting a result that is considered sufficient regarding the expected finding
- 1-3p. is given to results that are partly correct/insufficient regarding the expected finding
- 0p. is given for an incorrect/false result or not reporting the results before closing date

Scoring, round 2, 2017
The maximum score is **20** p**.**

The maximum score/specimen is as follows:
Specimen 001: 5 p.
Specimen 002: 5 p.
Specimen 003: 5 p.
Specimen 004: 5 p.

See Annex 3 for detailed scoring principles.

The experts of this round were Deputy Chief Physician Tapio Seiskari, M.D., Ph.D., Fimlab, Tampere, Clinical Microbiologist Antti Nissinen, Ph.D., Synlab and Chief Physician Antti Hakanen, M.D., Ph.D., TUCH Microbiology and Genetics.

End of report

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Annex 1. General Bacteriology, 2, 2017.

The MIC-results reported by the participating laboratories of specimen 001, *S. dysgalactiae* ssp. *equisimilis* ATCC® 12394™.

Antimicrobial agent	MIC-value (mg/L)	Inter-pretation	Reported results per followed standard			Results in all
			EUCAST	CLSI	CA-SFM	
Amikacin	<=8	R	1			1
Amoxicillin	0,016	S	1			
	0,03	S			1	
	<=0,25	S	1	1		4
Amoxicillin-clavulanate	<=4,2	S	1			1
Ampicillin	0,032	S		1		
	<=0,06	S		1		
	<0,25	S	1			
	<=0,25	S	1			
	0,25	S	1			
	8	S	1			6
Cefepime	<=0,5	S	1	1		2
Cefotaxime	0,03	S	1			
	<0,12	S	1			
	<=0,12	S	2	1		
	0,12	S	5			
	<0,25	S		1		
	<=0,5	S	1	1		
	0,5	S	1			
	546	S	1			15
Ceftriaxone	0,012	S	1			
	<0,12	S	2			
	<=0,12	S	1	1		
	0,12	S	5			10
Cefuroxime	<=0,25	S	1			1
Chloramphenicol	<=2	S	1	1		
	2	S			1	
	8	S	1			4
Clindamycin	0,06	S			1	
	0,0625	S	1	1		
	0,094	S		2		
	0,125	S	2			
	<0,25	S	3			
	<=0,25	S	3	1		
	0,25	S	4			
	<=0,5	S	1			
	0,5	S	6			
	0,5	R	1			26

Antimicrobial agent	MIC-value (mg/L)	Inter-pretation	Reported results per followed standard			Results in all
			EUCAST	CLSI	CA-SFM	
Doxycycline	0,94	S	1			1
Ertapenem	0,016	S		1		1
Erythromycin	0,047	S		2		
	<=0,0625	S		1		
	<0,12	S	2			
	<=0,12	S	5	1		
	0,12	S	6			
	0,125	S	2	1		
	0,19	S	1			
	0,25	S	2			
	0,38	S	1			
	<=0,5	S	1			
	0,5	S			1	
	>=8	R	1			
	546	S	1			28
Gentamicin	<=4	R	1			
	<250	S	1			
	250	S			1	3
Imipenem	0,047	S		1		1
Kanamycin	512	S	1			1
Levofloxacin	<0,25	S	1			
	<=0,25	S	5	1		
	0,25	S	6			
	<0,5	S	1			
	<=0,5	S	2	2		
	0,5	S	4	1	1	24
Linezolid	0,2	S	1			
	<=0,5	S		1		
	1	S	2		1	
	1,5	S	1			
	<2	S	1			
	<=2	S	4			
	2	S	6			
	4	S	1			18
Meropenem	0,016	S		1		
	<=0,06	S		1		
	<=0,125	S	1	1		3
Moxifloxacin	0,12	S	1			
	<=0,25	S	1			
	0,25	S	1			
	0,5	S	1			4

Antimicrobial agent	MIC-value (mg/L)	Inter-pretation	Reported results per followed standard			Results in all
			EUCAST	CLSI	CA-SFM	
Nitrofurantoin	<=32	S	1			
	546	S	1			2
Penicillin	0,006	S	1			
	<=0,012	S	2			
	0,012	S	1			
	0,016	S	2	1		
	0,023	S	2			
	<=0,03	S		1		
	0,03	S	1			
	<=0,0312	S	1	1		
	<=0,03125	S	2			
	<0,06	S	2			
	<=0,06	S	6	1		
	0,06	S	6		1	
	<0,25	S	1			
	546	S	2			34
Phosphomycin	<=32	R	1			
	32	R	1			2
Piperacillin-tazobactam	<0,5	S		1		1
Rifampicin	0,06	S	1			
	546	S	1			2
Streptomycin	<=1000	S	1			1
Teicoplanin	0,016	S	1			
	0,12	S	1			
	<1	S	1			
	<=1	S	1			
	1	S			1	
	<=2	S	1			
	546	S	1			7
Telithromycin	546	S	1			1
Tetracycline	0,125	S	1			
	<0,25	S	1			
	<=0,25	S	5	1		
	0,25	S	5			
	<0,5	S	1			
	<=0,5	S	1	2		
	0,5	S	3		1	
	>=16	R	1			
	546	S	1			23
Tigecycline	0,06	S	1			
	0,064	S	1			2

Antimicrobial agent	MIC-value (mg/L)	Inter-pretation	Reported results per followed standard			Results in all
			EUCAST	CLSI	CA-SFM	
Tobramycin	<=4	R	1			1
Trimethoprim-sulfamethoxazole	0,047	S	1			
	1	S	1			
	<=1,19	S	1			
	<10	S	1			
	<=10	S	5			
	10	S	4			
	19	S	1			14
Vancomycin	<0,12	S	2			
	<=0,12	S	2			
	0,12	S	5			
	0,19	S	2			
	0,25	S	1	1		
	<0,5	S	2			
	<=0,5	S	2	1		
	0,5	S	5	1	1	
	0,75	S		1		
	<=1	S	1			
	1	S		1		
	<2	S	1			
	4	S	1			30
n			223	41	11	275

ANNEX 2. General Bacteriology 1, round 2, 2017. Results of test kits, analyzers and mass spectrometry reported for specimens 003 and 004.

Specimen 003. *Candida glabrata*

Test kit or analyzer/result	In all	Result
API 20C AUX (bioMerieux)		
200040	1	*C. glabrata*
2000040	2	*C. glabrata*
ID 32 C (bioMerieux)		
0001000001	4	*C. glabrata*
MicroScan WalkAway (Beckman Coulter)		
046013000	1	*C. glabrata*
RapID Yeast Plus (Thermo Scientific)		
110006	1	*C. glabrata*
770016	1	*C. tropicalis*
VITEK 2 YST (bioMerieux)		
4000104000200100	2	*C. glabrata*
4000104000200110	1	*C. glabrata*
4000104000201110	1	*C. glabrata*
4010104000201111	1	*C. glabrata*
not reported	4	*C. glabrata*

Mass spectrometry MALDI TOF		
Microflex LT (Bruker Daltonik)		
1,72	1	*C. glabrata*
1,9	1	*C. glabrata*
1,98	1	*C. glabrata*
1,99	1	*C. glabrata*
2,00	1	*C. glabrata*
2,01	1	*C. glabrata*
2,06	2	*C. glabrata*
2,07	1	*C. glabrata*
2,08	1	*C. glabrata*
2,09	1	*C. glabrata*
2,11	1	*C. glabrata*
2,15	1	*C. glabrata*
2,16	1	*C. glabrata*
not reported	1	*C. glabrata*
VITEK MS (bioMerieux)		
99 %	1	*C. glabrata*
99,9 %	6	*C. glabrata*

Specimen 004. *Clostridium tertium*

Test kit or analyzer/result	In all	Result
ANAEROtest 23 (Erba Lachema)		
not reported	1	*C. ramosum* (53,7 %) ***/*** *C. tertium* (41,9 %)
API 20 A (bioMerieux)		
47746023	1	*C. tertium*
MicroScan WalkAway (Beckman Coulter)		
002012004	1	*Streptococcus mutans*
RapID ANA II (Thermo Scientific)		
005042	1	*Clostridium baratii*
275042	1	*C. tertium*
715002	1	no final result
Rapid ID 32 A (bioMerieux)		
4317000000	1	*C. tertium*
4713000000	1	*C. tertium*
4713004000	1	*C. tertium*
VITEK 2 ANC (bioMerieux)		
0417405150411	1	*C. tertium*
0417507142410	1	*C. tertium*
1416715172411	1	*C. tertium*
1417405172411	1	*C. tertium*
2407207132411	1	*C. tertium*
not reported	2	*C. tertium*
not reported	1	*Clostridium* sp.
not reported	1	*Sphingomonas paucimobilis*

Mass spectrometry MALDI TOF		
Microflex LT (Bruker Daltonik)		
2,0	1	*C. tertium*
2,1	1	*C. tertium*
2,265	1	*C. tertium*
2,3	2	*C. tertium*
2,35	1	*C. tertium*
2,37	1	*C. tertium*
2,42	4	*C. tertium*
2,43	1	*C. tertium*
2,53	1	*C. tertium*
not reported	2	*C. tertium*
VITEK MS (bioMerieux)		
99 %	1	*C. tertium*
99,9 %	7	*C. tertium*

PCR (in house)	1	*C. tertium*

ANNEX 3. General Bacteriology 1, round 2, 2017. Scoring summary.

Spec.	Finding	Species identification	Interpretation of the finding			Referral	Scores in all
			Significant pathogen	Possible pathogen	Non-significant pathogen, norm. flora	Would be sent forward	
001	***Streptococcus dysgalactiae* ssp. *equisimilis***	4p.	1p.				**5p.**
	***Streptococcus* sp., beta-hemolytic, group G**	4p.	1p.				**5p.**
	Streptococcus dysgalactiae ssp. d*ysgalactiae*	3p.	1p.				4p.
	Streptococcus sp., beta-hemolytic, group C	2p.	1p.				3p.
	Streptococcus sp., beta-hemolytic, group A	2p.	1p.				3p.
	Streptococcus sp., beta-hemolytic, group B	2p.	1p.				3p.
002	***Pseudomonas aeruginosa***	4p.	1p.				**5p.**
	Pseudomonas sp.	2p.	1p.				3p.
	Other findings (*Cedecea davisae)*						0p.
003	***Candida glabrata***	4p.	1p.				**5p.**
	Yeast / *Candida* sp. , other than *C. albicans*	2p.	1p.			1p.	4p.
	Candida sp.	2p.	1p.			1p.	4p.
	Candida tropicalis	1p.	1p.				2p.
004	***Clostridium tertium***	4p.	1p.				**5p.**
	Clostridium sp.	2p.				1p.	3p.
	Anaerobic, gram-positive (spore-forming) bacilli/rods	1p.				1p.	2p.
	Clostridium ramosum	1p.					1p.
	Other findings (*Streptococcus* sp., *Sphingomonas* sp.*)*						0p.