

# Microbiology EQA

Annual Workshop on External Quality Assurance Schemes in Cambodia

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- Exercises

# Pacific Paramedical Training Centre PPTC



**PACIFIC PARAMEDICAL  
TRAINING CENTRE**

HEALTH TECHNOLOGY FOR THE PACIFIC REGION



**WORLD HEALTH ORGANIZATION**

COLLABORATING CENTRE  
FOR EXTERNAL QUALITY ASSESSMENT IN HEALTH  
LABORATORY SERVICES

In 2014, **54 laboratories** participated in all or part of the program:

- 26 Pacific Island Countries
- 19 Cambodia**
- 4 Private Labs (Fiji)
- 3 Laos PDR
- 1 Timor Leste
- 1 Bhutan

**PPTC 2014 Microbiology  
37 labs**

**10 labs: Cambodia**

**PPTC 2018 Microbiology  
14 labs: Cambodia**

# EQA 2013 and 2017

2013/3/3  
 Urine  
 Identification: *Morganella morganii*  
 Your Result: *Morganella morganii*  
 Score: 3/3

*Overall Respondants results*

No. of labs with correct ID of *Morganella morganii* = 58%  
 No. of labs with ID of closely related species = 27%  
 No. of labs with incorrect ID or no ID = 15%

**Susceptibility Results:**

Antibiotic	Referee Result	Your Result
Ampicillin	Resistant*	Resistant*
Trimethoprim	Susceptible	
Nitrofurantoin	Resistant	Susceptible#
Norfloxacin	Susceptible	Susceptible
Gentamicin	Susceptible	Susceptible
Amox/Clavulanic Acid	Resistant*	Resistant*
Ciprofloxacin	Susceptible	Susceptible
Chloramphenicol	Resistant	
Cefuroxime	Resistant*	
Trimethoprim/Suphamethoxazole	Susceptible	Susceptible
Ceftriaxone	Susceptible*	Susceptible*
Imipenem	Susceptible	
Meropenem	Susceptible	

Nitrofurantoin- M. moragnii	2013 n=7	2017 n=13
Correct	2	10 correct, including 2 labs incorrect in 2013
Incorrect	2	1 (In 2013 did not test Nitrofurantoin)
Did not test	3	1
Wrong ID	0	1

# 2012 Programme

- 3 challenges
- 5 labs
- Participation: 100%

Lab1	Lab2	Lab3	Lab4	Lab5
87%	95%	93%	88%	100%

# Microbiology EQA Score

- 1 challenge 2017
- 3 challenges 2018
- 14 labs 2018
- Participation: 100%

Lab	2012	2017	2018
Lab participating	5	13	14
Score range	87-100	90-100	85-100
Average			
Rank			

# National AMR Surveillance: QA Indicator for GLASS

## 'Pathogens included in GLASS are covered by EQA'

3.1.2.1 Information on status of national AMR surveillance system  
 "indicators are monitored on a yearly basis to assess countries' progress"

	INDICATOR	
Quality Assurance (QA)	External Quality assurance (EQA) is provided for NRL	Yes/No/Not known
	EQA is performed for laboratory participating in GLASS	Yes/No/Not known
	EQA is covering bacterial identification and AST	Yes/No/Not known
	<u>Pathogens included in GLASS are covered by EQA</u>	Yes/Some/None/Not Known
	Type of AST standards followed	CLSI/EUCAST/Other

# How do PPTC Challenges meet the GLASS indicator?

	2017 National AMR Surveillance Pilot	2018 National AMR Surveillance SOP
AMR Surveillance	2017 EQA challenge pathogens	2018 EQA challenge pathogens
E.coli	0	1
S.aureus	1	2
K.pneumoniae	0	0
Salmonella	0	1
Acinetobacter	0	0
B. pseudomallei	0	0
S.pneumoniae	0	1
<b>QA indicator: Pathogens included in GLASS are covered by EQA</b>	<b>1 pathogen only (SOME)</b>	<b>4 pathogens (SOME)</b>



# 1. EQA tips and reminders



**PACIFIC PARAMEDICAL TRAINING CENTRE**

HEALTH TECHNOLOGY FOR THE PACIFIC REGION  
WHO COLLABORATING CENTRE FOR EXTERNAL QUALITY  
ASSESSMENT IN HEALTH LABORATORY SERVICES



**Pacific Paramedical Training Centre**  
**Microbiology Quality Assessment Programme**  
**July 2018**

**Programme 2018/2**

**Laboratory No: 047**

**Enter date received in Laboratory: 23/07/2018**

**Date results due at PPTC: 22/08/2018**

**Enter date results sent to PPTC: 20/08/2018**

**Read PPTC instructions carefully**

- Please work carefully and safely as these cultures could contain potential pathogens. Treat the specimens in accordance with the principles of good Laboratory Practice.
- Ensure that you make a copy of your results, as this report will not be returned to you.
- A result sheet is included for each specimen. Please ensure that it is filled in fully giving all the tests that you perform to identify the organism, including latex agglutination and anti-sera tests, and the results of these tests, (e.g. biochemical profile number).
- Fill in all columns of the antimicrobial susceptibility result sheet. Please give the full name of the antibiotic not just its abbreviation, record zone size, interpretation and whether or not you would report the antibiotic for the isolate.
- **Failure to record all your testing details may result in loss of marks.**
- Please ensure that your results are back at the PPTC by the date indicated. This is 4 weeks from the date of dispatch.
- We will send out the referee laboratory's results a few days after the due date.
- Please send your results back using the following forms.

I hope you enjoy this programme.

Warm regards,

Nicky Beamish  
Microbiology QA Co-ordinator



# All tests must be clearly recorded

- A result sheet is included for each specimen. Please ensure that it is filled in fully giving all the tests that you perform to identify the organism, including latex agglutination and anti-sera tests, and the results of these tests, (e.g. biochemical profile number).
- Fill in all columns of the antimicrobial susceptibility result sheet. Please give the full name of the antibiotic not just its abbreviation, record zone size, interpretation and whether or not you would report the antibiotic for the isolate.
- **Failure to record all your testing details may result in loss of marks.**

# 2018/3

- 2018/3/1: 6 / 14 labs grew *S. pneumoniae*
- 2018/3/2: All labs reported “No growth”

## **IMPORTANT CHANGE**

### **Reconstitution of Microbiology Samples (Lyophilised)**

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Where possible, process the samples in your biological safety cabinet (BSC).  
Observe all laboratory PPE protocols (wear laboratory coat, gloves etc.).

**For reconstitution, the PPTC recommends the following method:**

1. Use aseptic technics at all times.
2. Reconstitute one sample at a time to avoid contamination.
3. Place the vials in BSC, aseptically tear off the metal crimp cap and discard.
4. Aseptically remove the rubber stopper and place it on the BSC work area/ sterile bench surface.
5. Using a calibrated pipette, add **1ml** of nutrient broth provided with the samples to each of the Microbiology samples 1 to 3.
6. Recap the vial using the corresponding rubber stopper.
7. **Mix** the vial by **gently** swirling it 4 to 8 times.
8. Leave on the bench for **20mins** to allow complete reconstitution.
9. Use a sterile pipette to add a drop to appropriate media for culture, and streak the plate using a sterile loop.

**Culture immediately following the reconstitution process on appropriate agar plates and as per your laboratory protocol.**

**Storage:**

Before testing, the lyophilised samples must be kept in your specimen fridge at 2 - 8°C.  
Reconstituted sample can be stored in the fridge at 2 - 8°C for up to 4 weeks.

# EQA and Competency Testing

“Only one person may use EQA for competency during the EQA event”

“After the EQA event has been scored, EQA samples may be used to assess competency of others”



PRACTICAL GUIDANCE FOR  
CLINICAL MICROBIOLOGY



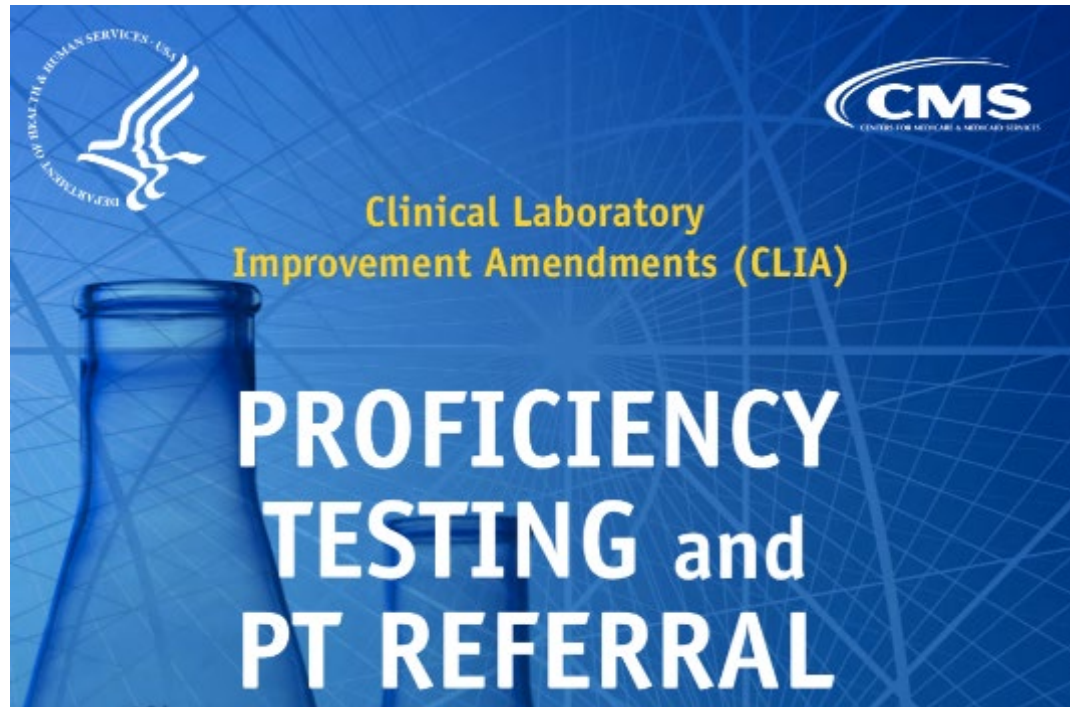
## Implementing a Quality Management System in the Medical Microbiology Laboratory

Roberta B. Carey,<sup>a\*</sup> Sanjib Bhattacharyya,<sup>b</sup> Sue C. Kehl,<sup>c</sup> Larissa M. Matukas,<sup>d</sup> Michael A. Pentella,<sup>e</sup> Max Salfinger,<sup>f</sup> Audrey N. Schuetz<sup>g</sup>

# Never discuss EQA (PT) before the cut-off date

## *May I discuss my PT results with another laboratory?*

**Never** discuss your PT results with another laboratory and **never** enter into discussion with another laboratory about their PT results before the PT event cut-off date. This activity may result in sanction(s) taken against your CLIA certificate.



# EQA 2013: lab responsibilities

- Continue full participation
- Treat the sample as a 'patient' sample
- Use resources available to you in your lab
- Do **NOT** refer isolates for further analysis
- Do **NOT** communicate with other labs about EQA
- Store isolates for follow-up
- Review: Result sheet, Immediate response, Individual response and laboratory worksheet
- Record and implement corrective actions when required
- Learn from both correct and incorrect results



# Support & Responsibilities

- Partners and MoH
  - Monitor EQA results, problems and corrective action
  - Regular meetings of NMMLN: EQA discussion, share experience
  - Ensure sufficient quality diagnostic supplies
  - Support equipment certification, maintenance and repairs
  - Provide training opportunities
- Hospital Leadership and PHD
  - Ensure sufficient staff
  - Ensure qualifications appropriate
  - Ensure sufficient quality diagnostic supplies
  - Consider laboratory manager recommendations
  - Support continuing education

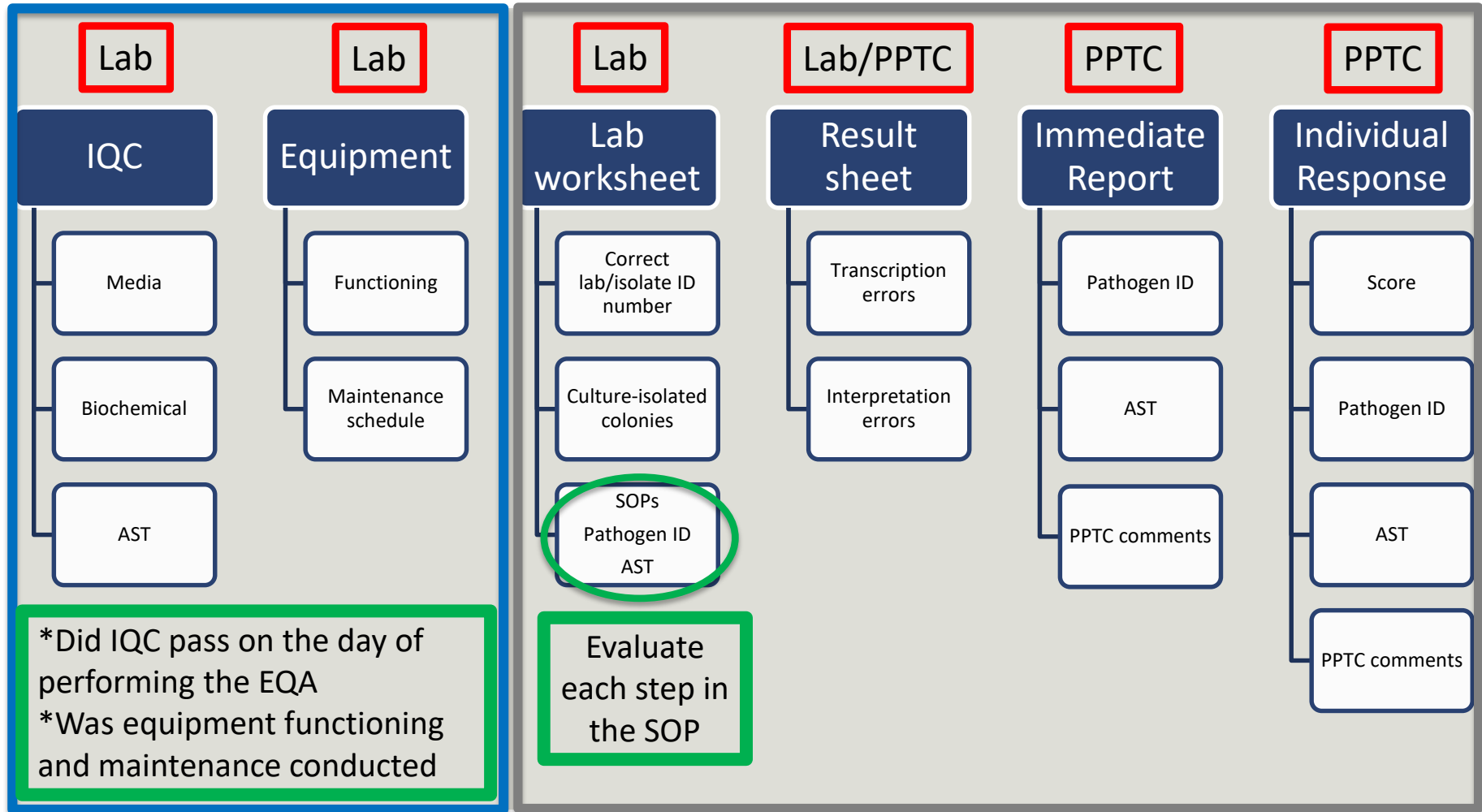


# What can labs do to correct problems

- All staff: read the Immediate and Individual response from PPTC
- Meeting
  - Review worksheets and IQC (Media, Biochemical, AST)
  - Discuss each step in the process
  - Identify possible sources of error
  - Investigate further with repeat testing (with IQC)
- Put a process in place to ensure the error is not repeated
- This may require review and revision of SOP

# EQA Exercise

# Systematic review process to detect problems and identify CA





# Exercise 1. 2018/1/1

Specimen 2018/1/1

This organism was isolated from a knee wound swab from an elderly man.

Identify the organism and perform antimicrobial susceptibility testing, if appropriate.

Use the result sheets below for reporting your identification of the organism and antimicrobial susceptibility results.

## Microbiology Result Sheet

- Culture Media you cultured the isolate onto: BAP, Mac, MSA and ASD
- Gram stain result: Gram positive cocci
- Identification Tests:  
In the chart below give the full name of the test performed and the result.  
**Please note - Failure to record these may result in loss of marks.**

TEST	RESULT
Catalase	Positive
Coagulase slide	Negative
Coagulase tube	Negative
Oxidase	Negative
PYR	Negative
Polymixin B	Resistance
Staphylococcus aureus latex	Negative

- Did you use an ID system (eg API)?                      Yes                       No
  - If yes, what ID system was it?
  - What was the profile number?
- Identity of the organism(s): Staphylococcus epidermidis

## Antibiotic Susceptibility Testing Result Sheet

Specimen 2018/1/1

Please fill in **all** the columns of this worksheet including full name of the antibiotic, not abbreviation, and whether or not your laboratory would report this antibiotic for this isolate in this clinical situation.

Antibiotic Name in FULL	Concentration of Disc	Disc expiry date	Test zone size	Interpretation R, S or I	Would you report this antibiotic? Yes or No
Cefotaxime	30ug	April 30 2020	6	R	No
Cloxacilline				R	Yes
Cefazoline				R	Yes
Chloramphenicol	30ug	Oct 31 2019	21	S	Yes
Clindamycin	2ug	Aug 31 2019	24	S	Yes
Erythromycin	15ug	Dec 31 2018	6	R	Yes
Penicillin	10unit	July 31 2018	6	R	Yes
Tetracycline	30ug	April 30 2021	21	S	Yes
Trimeth/Sulfa	1.25/23.75	July 31 2019	6	R	Yes
Note: MRSA detected					

# Exercise 2. 2017/1/1

Pacific Paramedical Training Centre

Microbiology Quality Assessment Programme

PROGRAMME 2017/1      INDIVIDUAL RESPONSE for Lab No. 040

This is an INDIVIDUAL response to the programme sent to you in March 2017. It gives the identification, and antimicrobial susceptibility results (where applicable), from the Reference Laboratories alongside your results for the organisms sent, plus any specific comments about your reply.

Please note:- There was an error in the immediate response sent in May for isolate 2017/1/3. The isolate is not susceptible to Clindamycin as previously reported. On repeat testing, using different methodology, the isolate was shown to have inducible resistance to Clindamycin.

The Vitek 2 instrument, which was used for initial and subsequent testing, of this isolate failed to detect this resistance mechanism. This is unusual as studies show good concordance for the Vitek 2 with manual disc diffusion methods.

Manual disc diffusion method (EUCAST) showed a clear D zone between Clindamycin and Erythromycin indicating inducible Clindamycin resistance.

## Summary of results from all replies:-

	2017/1/1	2017/1/2	2017/1/3
Correct to Species level	73%	27%	97%
Correct to Genus level	73%	94%	100%
Incorrect or inadequate identification	27%	6%	0%

# Exercise 2. 2017/1/1

## Antibiotic Susceptibility Testing Result Sheet **INDIVIDUAL RESPONSE** for Lab No. [REDACTED]

Specimen 2017/1/1 [REDACTED]

Please fill in all the columns of this worksheet including full name of the antibiotic, not abbreviation, and whether or not your laboratory would report this antibiotic for this isolate in this clinical situation.

Antibiotic Name in FULL	Concentration of Disc (µg)	Disc expiry date	Test zone size	Interpretation R, S or I	Would you report this antibiotic? Yes or No
Ampicillin (AM)	10	30/06/2018	6	R	Yes
AmoxiClav (AMC)	30	28/02/2017*	6	R	Yes
Cefazolin (CZ)	30	30/04/2019	6	R	Yes
Ceftriazone (CRO)	30	30/06/2018	30	S	Yes ( CZ= R )
Ceftazidime (CAZ)	30	31/12/2017	28	S	No
Cefepime ( FEP)	30	30/04/2018	26	S	No
Ciprofloxacin (CIP)	5	31/08/2019	29	S	Yes
Imipenem (IMP)	10	30/09/2018	24	S	No
Meropenem (MEM)	10	28/02/2018	29	S	No (Ceftri=S)
Gentamicin (GM)	10	30/06/2020	23	S	Yes
Amikacin (AN)	30	31/07/2018	24	S	No (G: S)
Fosfomycin (FOS)	200	31/07/2017	6	R	yes
Nitrofurantoin ( FT)	300	29/02/2020	15	I	Report Resistant
Trim/Sulfa (SXT)	1.25/23.75	31/07/2019	24	S	Yes
ESBL Producer? Y/N					<b>No Produce</b>

2017/1/1

Urine

Identification:

*Morganella morganii*

Your result:

*Morganella morganii*

Score:

3/3

Susceptibility Results:

Antibiotic	Referee result	Your result
Ampicillin	Resistant	Resistant
Amoxicillin/Clavulate	Resistant	Resistant
Trimethoprim	Susceptible	Not tested
Nitrofurantoin	Resistant	Intermediate*
Cotrimoxazole	Susceptible	Susceptible
Ciprofloxacin	Susceptible	Susceptible
Norfloxacin	Susceptible	Not tested
Gentamicin	Susceptible	Susceptible
Ceftazidime	Susceptible	Susceptible
Ceftriaxone	Susceptible	Susceptible
Meropenem	Susceptible	Susceptible

Score:

8/9



Pacific Paramedical Training Centre  
Microbiology Quality Assessment Programme

# Exercise 3: 2017/1/3

PROGRAMME 2017/1      INDIVIDUAL RESPONSE for Lab No. 040

This is an INDIVIDUAL response to the programme sent to you in March 2017. It gives the identification, and antimicrobial susceptibility results (where applicable), from the Reference Laboratories alongside your results for the organisms sent, plus any specific comments about your reply.

Please note:- There was an error in the immediate response sent in May for isolate 2017/1/3. The isolate is not susceptible to Clindamycin as previously reported. On repeat testing, using different methodology, the isolate was shown to have inducible resistance to Clindamycin.

The Vitek 2 instrument, which was used for initial and subsequent testing, of this isolate failed to detect this resistance mechanism. This is unusual as studies show good concordance for the Vitek 2 with manual disc diffusion methods.

Manual disc diffusion method (EUCAST) showed a clear D zone between Clindamycin and Erythromycin indicating inducible Clindamycin resistance.

## Summary of results from all replies:-

	2017/1/1	2017/1/2	2017/1/3
Correct to Species level	73%	27%	97%
Correct to Genus level	73%	94%	100%
Incorrect or inadequate identification	27%	6%	0%

# Exercise 3: 2017/1/3

## INDIVIDUAL RESPONSE for Lab No. ██████

2017/1/3

Nasal Swab

Identification:

*Staphylococcus aureus*\*

Your result:

*Staphylococcus aureus*

Score:

3/3

### Susceptibility Results:

<i>Antibiotic</i>	<i>Referee result</i>	<i>Your result</i>
Penicillin	Resistant	Not tested
Flucloxacillin (Cefoxitin)	Resistant	Resistant
Erythromycin	Resistant	Resistant
Tetracycline/Doxycycline	Resistant	Susceptible
Cotrimoxazole	Resistant	Resistant
Gentamicin	Resistant	Not tested
Ciprofloxacin	Susceptible/Resistant	Not tested
Clindamycin	Resistant #	Susceptible
Vancomycin	Susceptible	Susceptible

**\*This isolate is an MRSA.**

**#This isolate showed inducible resistance to Clindamycin.**

*Inducible clindamycin resistance can be detected by antagonism of clindamycin activity and a macrolide agent. Place the erythromycin and clindamycin disks 12-20 mm apart (edge to edge) and look for antagonism (the D phenomenon). For more detail see:-*

[http://www.eucast.org/fileadmin/src/media/PDFs/EUCAST\\_files/Disk\\_test\\_documents/Version\\_5/Reading\\_guide\\_v\\_5.0\\_EUCAST\\_Disk\\_Test.pdf](http://www.eucast.org/fileadmin/src/media/PDFs/EUCAST_files/Disk_test_documents/Version_5/Reading_guide_v_5.0_EUCAST_Disk_Test.pdf)

Score:

4/6

# Exercise 3: 2017/1/3

## Result Sheet

Specimen 2017/1/3

Laboratory No: XXXXXXXXXX

This organism was isolated from a nasal swab of a child with recurrent boils.

Identify the organism and perform antimicrobial susceptibility testing, if appropriate.

Use the result sheets below for reporting your identification of the organism and antimicrobial susceptibility results.

### Microbiology Result Sheet

- Culture Media you cultured the isolate onto:

**MSA, BAP, MAC**

- Gram stain result:.....**Gram positive cocci** .....

- Identification Tests:

In the chart below give the full name of the test performed and the result.  
Please note - Failure to record these may result in loss of marks.

TEST	RESULT
Catalase	Positive
Slide coagulase	Positive
Tube coagulase	Positive

- Did you use an ID system (eg API)? Yes No
  - If yes, what ID system was it? .....
  - What was the profile number? .....
- Identity of the organism(s): **Staphylococcus aureus**

### Antibiotic Susceptibility Testing Result Sheet

Specimen 2017/1/3

Laboratory No: XXXXXXXXXX

Please fill in all the columns of this worksheet including full name of the antibiotic, not abbreviation, and whether or not your laboratory would report this antibiotic for this isolate in this clinical situation.

Antibiotic Name in FULL	Concentration of Disc	Disc expiry date	Test zone size	Interpretation R, S or I	Would you report this antibiotic? Yes or No
Cefoxitin (FOX)	30ug	31-10-2017	06	Resistant	<b>No. Report Oxacillin as "Resistant"</b>
Erythromycin (E)	15ug	30-11-2019	06	Resistant	<b>Yes</b>
Clindamycin (CC)	2ug	30-04-2017	21	Susceptible	<b>Yes</b>
Trimethoprim-Sulfamethoxazole (SXT)	1.25ug/23.75ug	31-01-2018	10	Resistant	<b>Yes</b>
Doxycycline (D)	30ug	26-02-2018	17	Susceptible	<b>Yes</b>
Vancomycin (VAN)	(E-test)	19-10-2020	MIC = 1,5ug/ml	Susceptible	<b>Yes</b>
Rifampin (RIF)	5ug	28-02-2018	32	Susceptible	<b>Yes</b>

# Thank you

