September 2020

INSTAND

Manual

Manual for Qualification Control Testing of Bacteriology No. 412

The products must be handled like patient samples!

DAkkS
Deutsche
Akkreditierungsstelle
D-EP-15027-02-00

Modifications in the EQAS 412

valid from	Modification
A2-2019	Online entry of results via the following link: https://www.instand-ev.de/ringversuche-online.html; revision of this manual
A1-2019	Coding of the intermediate category after renewed definition by EUCAST and EUCAST+NAK resp. in the susceptibility testing. Revision of the code tables and the corresponding user instructions.
A2-2018	New revision of the note on the renewed taxonomy of Enterobacteriaceae/ Enterobacterales
A1-2018	Revision of the note on the renewed taxonomy of Enterobacteriaceae/ Enterobacterales
A1-2017	Note on the renewed taxonomy of Enterobacteriaceae/ Enterobacterales
	Note on inferred categorized results in susceptibility testing
A1-2017	Link for the website of the results updated
A2-2016	Revision of code tables, especially code table 6
A1-2016	Susceptibility testing allowed using supplemental breakpoints according NAK Deutschland (www.NAK-Deutschland.org), in addition to CLSI and EUCAST Revision of code tables, especially code table 4
A2-2015	code „3“ = „evaluation not possible“ in reporting of susceptibility results discarded
A1-2014	Susceptibility testing allowed according to CLSI or EUCAST (www. EUCAST.org) only
A1-2014	Gram staining and related new code tables (code 7 to 9)

INSTAND

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Results available as of **2020-09-28**
at www.instand-ev.de

EQAS in Bacteriology A2/2020 incl. Gram staining

Dear colleague,

we are sending you herewith 5 samples for the external quality assessment schemes. The samples consist of lyophilized strains. Therefore we are able to provide anaerobic or microaerophilic strains. Please consider this when you select media and incubation conditions for reconstitution of the strains. The samples must be rehydrated and cultured according to the guideline (see enclosed "Supplement").

Sources of samples:

Sample 1: midstream urine
Sample 2: midstream urine
Sample 3: wound swab from ulcer
Sample 4: blood culture
Sample 5: blood culture - **no susceptibility testing requested!** -

The targets of this survey are:

1. Gram staining of the strains. In case of successful participation a certificate will be issued.

2. Identification of the strains

3. Susceptibility testing according to the chosen susceptibility testing standard (CLSI, EUCAST (www.EUCAST.org)). Reporting of the used standard is obligatory.
   Susceptibilty testing according to modified breakpoints published by NAK, www.NAK-Deutschland.org, is recommended for participants from Germany to gain consistency with approvals for antiinfective substances by the German Bundesinstitut für Arzneimittel und Medizinprodukte (BfArM).
   Please mark the applied standard:
   - CLSI
   - EUCAST (www.EUCAST.org)
   - EUCAST + NAK (www.NAK-Deutschland.org)

4. Finishing results until the given deadline.

Additional instructions:

1. With immediate effect online entry of results take place. Please use the following website https://www.instand-ev.de/en/eqas-online/ordering-online-and-entering-results/ for documentation of the results. You will find short instructions `RV-Online Manual´ and `Quick Guide Online Ordering´. A result sheet for internal use is provided in the area `Result Entry´.
   After the results have been entered, you can use the "Print Preview" button to print out your results or save them in PDF format. We recommend that you print out / save each EQAs as proof of the timely input of your values.

2. Antibiotics to be tested should be selected according to gram staining results, i.e. either the antibiotic panel for gram positive bacteria or gram negative bacteria has to be selected. In addition, please note:

   a. Each participant is responsible for the selection of the appropriate antibiotic panel

   b. Only results defined by the susceptibility testing standard used should be reported

   c. In any case all antibiotics with given breakpoints by the respective susceptibility testing standard are rateable. Moreover, in several cases additional antibiotics without given breakpoints are rateable by inferring the categorical results from other, prototypic substances. Target values will be set for inferred ratings too. Examples with respect to e.g. EUCAST regarding the antibiotics requested in the bacteriology EQA are:

Species or group	rated antibiotic substance	rating inferred for
Staphylococcus aureus u. *lugdunensis*	Penicillin	Ampicillin
Staphylococcus spp.	Cefoxitin-Screen oder Oxacillin	Oxacillin, Cefuroxime
Streptococcus Gr. A, B, C und G	Penicillin	Ampicillin, Cefuroxime

   The minimal number of antibiotics to be tested will be determined by the antibiotics with available breakpoints. Reporting of inferred results is reasonable, as with a higher number of results the impact of false results will be diminished.
   At least 6 of the 8 antibiotics have to be tested. If, according to the used standard, less than 6 antibiotics are evaluable, all of them have to be reported.

   d. Evaluation of results will be based on the version of standards valid at the date of EQAS sending.

e. The possible results for susceptibility testing are:

- resistant
- intermediate (CLSI) or susceptible at increased exposure (EUCAST +/- NAK)
- susceptible
- not performed

The coding considers the renewed definition for the "intermediate" category as "susceptible at high exposure" by EUCAST, introduced in 2019. For further information read respective documents on the EUCAST website (www.eucast.org).

Antibiotic panels:

Antibiotic substances	Gram positive bacteria	Gram negative bacteria
Penicillin	Penicillin	-
Ampicillin	Ampicillin	Ampicillin
Piperacillin-tazobactam	-	Piperacillin-tazobactam
Oxacillin	Oxacillin	-
Cefuroxime	Cefuroxime	Cefuroxime
Ceftazidime	-	Ceftazidime
Meropenem	-	Meropenem
Gentamicin	Gentamicin	-
Tobramycin	-	Tobramycin
Ciprofloxacin	Ciprofloxacin	Ciprofloxacin
Erythromycin	Erythromycin	-
Vancomycin	Vancomycin	-
Trimethoprim/Sulfamethoxazol	-	Trimethoprim/Sulfamethoxazol

3. Since December 2016 the bacteria formerly grouped in the family "Enterobacteriaceae" are now grouped under the new order "Enterobacterales". The EUCAST standard considers these changes since 2018. CLSI still does not consider these changes in 2019. Susceptibility testing based on the CLSI standard for respective bacteria should be performed using the CLSI tables for Enterobacteriaceae.

4. The task will not be rated as solved if the closing date for submitting the results is passed

5. Only one result in each field is allowed, with the exception of the field `Appraisal / classification / IFSG reporting obligation (Germany only)´, which allows multiple entries.

6. Please report results strictly according to the numbering on the samples. Erroneous assignments will not be corrected.

7. Please enter results at the latest on **2020-09-18** on the following website https://www.instand-ev.de/en/eqas-online/ordering-online-and-entering-results/

8. You should keep the strains until receipt of EQAS results for retesting in case of wrong results.

9. After the deadline, beginning **2020-09-28** results for the five samples will be provided by Internet: http://www.instandev.de/en/news/.

10. Questions concerning

    a. shipping of the external quality assessment samples and newly introduced online entry should be addressed to Instand e.V.,

    b. processing of the samples or certificates should be addressed to the institute of the head of EQAS, phone +49-511-532-4368.

Good luck and thank you for your cooperation

Best regards

Prof. D. Schlüter
Adviser

OA Dr. med. S. Ziesing
Assistant Adviser

SUPPLEMENT

Rehydration and Culture of the Microbial Strains

The cultures to be examined were lyophilized. In order to maintain the viability of the cultures the vials should not be opened and kept in the refrigerator, better in a deep-freezer, until they are used. The vials were closed under vacuum following production. Therefore the generation of aerosols while removing the rubber stoppers cannot be excluded. Vials should be opened and processed within a safety cabinet wearing gloves. Additionally a cloth, drained with disinfectant should be held over the vials while removing the rubber stopper.

To avoid cross contaminations, each strain should be opened, reconstituted and plated separately.

For the culturing of the strains the following procedure is recommended:,

2. Remove vials from package.

3. Remove screw caps.

4. Take the rubber stoppers from the vials using a pair of tweezers.

5. Reconstitute the lyophilisates by adding 1 ml of a non-selective culture broth (i.e. trypticase soy broth).
6. Dissolve the lyophilisates completely. Plate out one drop of each suspension onto suitable, non-selective agar media, including media for anaerobic and microaerophilic culture. Incubate the cultures at 36° C.
7. Transfer the remainder of each suspension into tubes with 5 ml of a suitable broth medium and incubate for 1 or more days at 36° C aerobic and anaerobic. Only when solid media show no growth, an agar medium has to be inoculated from the broth culture.