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Original article

The quality of antimicrobial discs from nine manufacturers—EUCAST evaluations in 2014 and 2017

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ABSTRACT

Objectives: Antimicrobial discs for susceptibility testing can be obtained from many manufacturers. We evaluated the quality of discs from nine manufacturers in 2014 and 2017.

Methods: Antimicrobial discs of 16 agents from nine manufacturers were evaluated using EUCAST criteria. Discs were tested in triplicate on Müller–Hinton medium against EUCAST quality control (QC) strains. Mean values were compared with targets and ranges in the EUCAST QC tables.

Results: Three manufacturers (Becton Dickinson, Mast and Oxoid) demonstrated excellent and consistent disc quality both in 2014 and 2017. Manufacturers with discs of inadequate quality improved their results between the two periods. Overall, 92% (795/861) versus 97% (1038/1071) of zone diameter readings were within QC ranges and 58% (497/861) versus 75% (806/1071) were within the QC target ± 1 mm, for the first and second studies, respectively. One manufacturer (HiMedia) had major quality problems with 33% (26/78) of readings out of range in the first study and 17% (20/120) in the second study. Discs from some manufacturers showed unexpected variation in inhibition zone diameters (4–9 mm) for discs within the same vial.

Conclusions: Antimicrobial discs from three of nine manufacturers exhibited excellent and reproducible quality. The discs of the other six manufacturers demonstrated various quality issues, some of which were severe. After presenting the results to manufacturers and users, all managed to improve the quality. Our study points to the need for more stringent criteria for disc manufacturing. Criteria should not only address the nominal potency of discs but also define the end result. **J. Åhman, Clin Microbiol Infect 2019;25:346**

Introduction

Disc diffusion is used worldwide for antimicrobial susceptibility testing (AST). The method is simple, flexible and inexpensive, traits that have become increasingly important with the introduction of new agents, where the slow implementation in automated AST systems has become a problem [1]. However, users and manufacturers may consider the simplicity and flexibility of the method a carte blanche for ignoring the need for stringent quality control (QC).

In 2009, EUCAST (the European Committee on Antimicrobial Susceptibility Testing) published a standardized disc diffusion method [2] calibrated to EUCAST clinical MIC breakpoints. Parameters such as preparation of media, inoculum preparation, inoculation of plates, incubation conditions, reading of results and QC criteria for users and manufacturers were detailed. Correct and reproducible results will be obtained only when laboratories use materials of good quality, adhere to the methodology and conduct rigorous quality control. In-house practices concerning storage and handling will influence the end-user quality of materials, but the primary responsibility for product quality is with the manufacturers. However, the final responsibility for the correctness of AST results rests with the individual laboratory. The standards for production of antimicrobial discs available to manufacturers, the US Food and Drug Administration [3], Deutsches Institut für Normung [4] and the World Health Organization [5], only address the issue of the nominal potency with limits of acceptable mean disc content being 67%–150%, 90%–125% and 75%–135% of the declared content, respectively.

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In the present study, we aimed to identify all manufacturers of antimicrobial discs and evaluate the performance of a large number of relevant antimicrobial discs. To the best of our knowledge this has not been done before, although there are published comparisons of discs from some manufacturers [6–8] and articles criticizing the manufacturing quality of discs [9–11]. The performance of antimicrobial discs is evaluated by repeat testing of type culture collection strains (QC strains) [12], for which there are defined [13] inhibition zone diameter targets to control accuracy, and ranges to evaluate precision. Repeat testing of these strains should result in inhibition zone diameters close to the target value with occasional variation within the range. The present study was performed on two occasions (2014 and 2017) and following the first study, results were presented to the manufacturers and users. We wanted to see if such a procedure could help to improve the quality of discs.

Materials and methods

Sixteen antimicrobial discs were selected to represent different antibiotic classes and to include screening discs for important resistance mechanisms (Table 1). Discs were obtained on two occasions (in 2014 and 2017) from nine international manufacturers: Abtek Biologicals Ltd (Liverpool, UK), Becton Dickinson (BD, Sparks, MD, USA), Bioanalyse (Ankara, Turkey), Bio-Rad (Marnes-la-Coquette, France), HiMedia (Mumbai, India), Liofilchem (Roseto degli Abruzzi, Italy), Mast Diagnostics (Bootle, UK), Oxoid/Thermo Fisher Scientific (Basingstoke, UK) and SirScan/i2a Diagnostics (Montpellier, France). The discs were transported and stored according to the instructions of the manufacturers.

All tests were performed according to EUCAST disc diffusion methodology [12] at the EUCAST Development Laboratory. Discs were tested against QC strains (one to three per agent, Table 2) recommended by EUCAST and CLSI on in-house produced Müller–Hinton (MH) and MH-F (MH with 5% defibrinated horse blood and 20 mg/L β-NAD) agar plates. Agar from Oxoid (Thermo Fisher Scientific) was used for all discs, except for aminoglycosides, where MH agar from Becton Dickinson (BBL™) was used to avoid a known problem when testing aminoglycosides on Oxoid MH agar. For agents where there was only one suitable QC strain, the disc was investigated on both Oxoid and BBL agar (second study). Each combination of agent and QC strain was tested in triplicate. This resulted in a maximum of 102 readings per manufacturer in the first study and 120 readings in the second study (Table 2). Discs used for the triplicate tests were always from the same lot (see Supplementary material, Table S1) and the same vial. All triplicate tests were performed on the same day using three individually prepared inoculum suspensions. For each agent, one disc from each manufacturer was placed on the same 140-mm circular agar plate (examples in Fig. 1), to minimize variations due to differences in inoculum size, medium and incubation conditions. As a result of the large inhibition zones for meropenem, it was necessary to use two plates to avoid interference between zones. Zone diameters were measured to the nearest millimetre with a calliper.

In the second study, two QC strains were added to test the inhibitor component of β-lactam–β-lactamase inhibitor combination discs (criteria not available during the first study): *Escherichia coli* ATCC 35218 (TEM-1 β-lactamase-producer) for amoxicillin-clavulanic acid and *Klebsiella pneumoniae* ATCC 700603 (SHV-18 extended spectrum β-lactamase producer) for piperacillin-tazobactam.

Disc-to-disc variation

As part of the second study, the disc-to-disc variation in potency between discs within a vial was investigated for the following agents: amoxicillin-clavulanic acid, piperacillin-tazobactam and tobramycin. For each agent, 20 consecutive discs from one vial were investigated simultaneously using the same inoculum suspension. The three discs were tested with *E. coli* ATCC 25922, and the combination discs were tested also with the β-lactamase-producing QC strains *E. coli* ATCC 35218 and *K. pneumoniae* ATCC 700603. The test was repeated for all nine manufacturers and all results were read by one technician.

Data analysis

Mean values from triplicate tests of each disc were compared with QC target values and ranges in the EUCAST QC Tables version 7.0, 2017 [13], for the respective QC strain. Preliminary results from the first study were reanalysed because a few QC criteria (three of 33, see Supplementary material, Table S2) changed between 2014 and 2017. The result for each disc was categorized and each category was represented by a colour. The categorization depended on how the mean value of the three tests related to the QC criteria: within-target ±1 mm (green), >1 mm but within ±2 mm from target (yellow), >2 m from target but within the QC range (orange) or if outside the QC range (red). When the categorization differed between QC strains, the dominant categorization was chosen to describe the quality of the disc. It was also noted whether the mean values were above (High) or below (Low) the targets. Discs with mean values within range but with a single reading out of range were highlighted in tables. When a variation of ≥4 mm for triplicate

Table 1
Rationale for selection of antimicrobial discs

Antimicrobial disc	Criteria for inclusion
Benzylpenicillin 1 unit	Screen for β-lactam non-susceptibility in *Haemophilus influenzae*
Amoxicillin-clavulanic acid 20–10 μg	Clinically important and often used agent
Piperacillin-tazobactam 30–6 μg	Clinically important and often used agent
Oxacillin 1 μg	Screen for β-lactam non-susceptibility in *Streptococcus pneumoniae.*
Mecillinam 10 μg	Important because broth microdilution not possible
Cefotaxime 5 μg	Clinically important and often used agent. Important for detection of extended spectrum β-lactamase in *Enterobacteriaceae*
Cefoxitin 30 μg	Screen for methicillin resistance in staphylococci
Ceftazidime 10 μg	Clinically important and often used agent. Important for detection of extended spectrum β-lactamase in *Enterobacteriaceae.*
Meropenem 10 μg	Screen for carbapenem resistance in *Enterobacteriaceae* and for detection of carbapenemase-producing *Enterobacteriaceae*
Ciprofloxacin 5 μg	Clinically important and often used agent. Represents fluoroquinolones
Pefloxacin 5 μg	Screen for fluoroquinolone resistance including low-level resistance in *Salmonella* spp.
Norfloxacin 10 μg	Screen for fluoroquinolone resistance including low-level resistance in Gram-positive organisms
Gentamicin 10 μg	Clinically important and often used agent
Tobramycin 10 μg	Clinically important and often used agent
Erythromycin 15 μg	Important agent used for screening for all macrolide resistance
Tetracycline 30 μg	Clinically important and often used agent. Used as screen for tetracycline resistance

Table 2
Quality control strains and Müller–Hinton agar included per antimicrobial agent; each combination was tested in triplicate

Antimicrobial agent	Quality control strains								MH agar		Max no of tests per manufacturer	
	E. coli ATCC 25922	*P. aeruginosa* ATCC 27853	*S. aureus* ATCC 29213	*En. faecalis* ATCC 29212	*Str. pneumoniae* ATCC 49619	*H. influenzae* ATCC 49766	*E. coli* ATCC 35218	*K. pneumoniae* ATCC 700603	Oxoid	BBL	1st study	2nd study
Benzylpenicillin 1 unit			●		●	●			X		9	9
Amoxicillin-clavulanic acid 20–10 μg	●						●[a]		X	X[a]	3	9
Piperacillin-tazobactam 30–6 μg	●	●						●[a]	X		6	9
Oxacillin 1 μg			●[a]		●				X		3	6
Mecillinam 10 μg	●								X	X[a]	3	6
Cefotaxime 5 μg	●				●	●			X		9	9
Cefoxitin 30 μg			●						X	X	6	6
Ceftazidime 10 μg	●	●							X		6	6
Meropenem 10 μg	●	●			●				X		9	9
Ciprofloxacin 5 μg	●		●	●					X		9	9
Pefloxacin 5 μg	●								X	X[a]	3	6
Norfloxacin 10 μg	●		●	●					X		9	9
Gentamicin 10 μg	●		●							X	6	6
Tobramycin 10 μg	●		●							X	6	6
Erythromycin 15 μg			●		●				X		6	6
Tetracycline 30 μg			●		●	●			X		9	9
Total											102	120

E. coli, *Escherichia coli*; *P. aeruginosa*, *Pseudomonas aeruginosa*; *S. aureus*, *Staphylococcus aureus*; *En. faecalis*, *Enterococcus faecalis*; *Str. pneumoniae*, *Streptococcus pneumoniae*; *H. influenzae*, *Haemophilus influenzae*; *K. pneumoniae*, *Klebsiella pneumoniae*.
[a] Only included in second study.

tests was found and confirmed, the overall result for that disk was categorized as unacceptable (red colour) and highlighted in tables, even if the mean value was within range.

The number of individual zone diameter readings within QC ranges and on QC target ±1 mm was calculated for each disc and manufacturer. For each manufacturer, the total percentage of out-of-range results was also calculated.

Ethical considerations

There were no patient strains involved in this study. All testing was performed *in vitro* using type culture collection strains.

Results

Of the total number of disc lots (nine manufacturers, 16 discs; $n = 144$) requested from the manufacturers, the number of supplied lots was 135 (94%) in the first study and 143 (99%) in the second study, resulting in a total number of 861 versus 1071 inhibition zone diameter readings, respectively (Fig. 2). The combined results for all strains tested per agent and manufacturer are shown in Fig. 2, for both studies. Of the total number of zone diameter readings in the first and the second study, 92% (795/861) versus 97% (1038/1071) were within QC ranges and 58% (497/861) versus 75% (806/1071) were within QC targets ±1 mm. In the first study, >99% (305/306) of the readings for discs from Becton Dickinson, Mast and Oxoid were within the QC ranges. In the second study, all readings were within the QC ranges for all discs from these three manufacturers plus those from Bioanalyse, Bio-Rad and SirScan.

In the first study, out-of-range results were mainly observed for discs from HiMedia, Abtek, Liofilchem and Bio-Rad with 33% (26/78), 11% (9/84), 10% (10/102) and 7.3% (7/96) of readings out-of-range, respectively. Discs from HiMedia often produced inhibition zones that were too large, with some results up to 10 mm above the

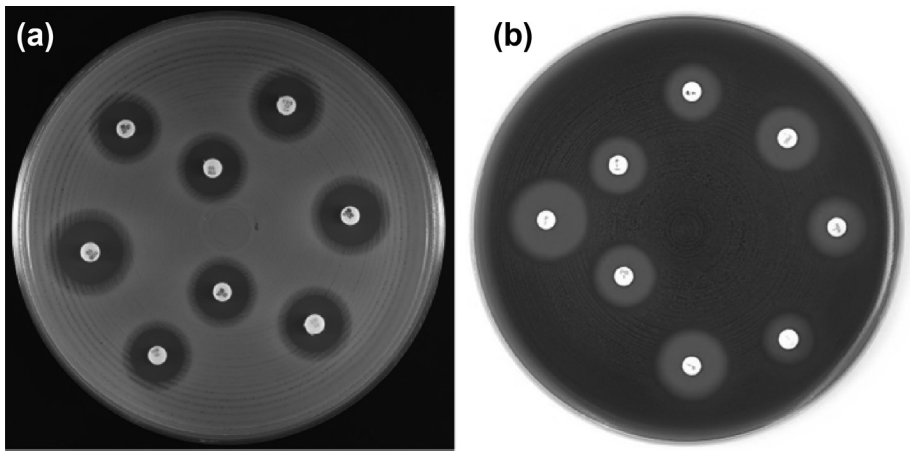

Fig. 1. Activity of equipotent discs from different manufacturers. (a) Tobramycin 10-μg discs versus *Escherichia coli* ATCC 25922; (b) Benzylpenicillin 1-unit discs versus *Streptococcus pneumoniae* ATCC 49619.

STUDY 2014

Antimicrobial disk	Abtek	BD	Bio-analyse	Bio-Rad	HiMedia	Liofil-chem	Mast	Oxoid	SirScan	
Benzylpenicillin 1 unit		L	H		NA				H	
Amoxicillin-clav. 20-10 µg	L			H	H					
Piperacillin-tazo. 30-6 µg	L				NA				H	
Oxacillin 1 µg		L	L		H	L			L	
Mecillinam 10 µg	L		H		H				H	
Cefotaxime 5 µg[1]	NA				NA					
Cefoxitin 30 µg	NA			H*	L*	H				
Ceftazidime 10 µg	L				L**					
Meropenem 10 µg[1]	L			H	H	H*		H		
Ciprofloxacin 5 µg[1]		L			H		L			
Pefloxacin 5 µg	NA	L			H	L			NA	
Norfloxacin 10 µg	L				H*				L	
Gentamicin 10 µg		H			H				NA	
Tobramycin 10 µg				NA	H*	H				
Erythromycin 15 µg	L	L	L*		H	L			L	
Tetracycline 30 µg	L	L*	L			L	L		L*	
Total number of readings	84	102	102	96	78	102	102	102	93	**861**

STUDY 2017

Antimicrobial disk	Abtek	BD	Bio-analyse	Bio-Rad	HiMedia	Liofil-chem	Mast	Oxoid	SirScan	
Benzylpenicillin 1 unit		L							NA	
Amoxicillin-clav. 20-10 µg	L			H	L**	L				
Piperacillin-tazo. 30-6 µg	L		H		L**		H			
Oxacillin 1 µg				H	H					
Mecillinam 10 µg										
Cefotaxime 5 µg										
Cefoxitin 30 µg		L	H		L					
Ceftazidime 10 µg	L				L*					
Meropenem 10 µg					L*					
Ciprofloxacin 5 µg										
Pefloxacin 5 µg					H	L				
Norfloxacin 10 µg										
Gentamicin 10 µg					H		H			
Tobramycin 10 µg					H					
Erythromycin 15 µg										
Tetracycline 30 µg					L					
Total number of readings	120	120	120	120	120	120	120	120	111	**1071**

[1]Data reanalysed due to changes in QC criteria since 2014.

Mean value within ± 1 mm of the target value
Mean value >1 mm but within ± 2 mm of the target value
Mean value >2 mm from target value but still within the QC range
Mean value out of the QC range

NA = Not Available
H = High, mean value > 1 mm above target
L = Low , mean value > 1 mm below target
* Single reading out of QC range
** Variation ≥4 mm for consecutive tests

Fig. 2. Results for discs from nine manufacturers versus EUCAST quality control targets and ranges.

QC target values. In the second study, out-of-range results were observed for discs from HiMedia, Liofilchem and Abtek with 17% (20/120), 8.3% (10/120) and 2.5% (3/120) of readings being out-of-range, respectively.

The following discs were categorized as unacceptable (red) because of a confirmed variation of ≥4 mm for consecutive tests: ceftazidime discs from HiMedia in the first study and both amoxicillin-clavulanic acid and piperacillin-tazobactam discs from HiMedia in the second study.

Disc-to-disc variation

The large variation observed for the β-lactam–β-lactamase inhibitor combination discs from one manufacturer in the second study led us to further investigate this. Amoxicillin-clavulanic acid and piperacillin-tazobactam, plus tobramycin, for which we had previously obtained stable results, were selected for this investigation. The results are presented in Table 3. A variation in inhibition zone diameters of ≤3 mm between discs from a single vial was observed for tobramycin discs from all manufacturers (examples in Fig. 3a,b). However, the disc-to-disc variation was large both for amoxicillin-clavulanic acid and piperacillin-tazobactam discs from HiMedia, with zone diameters varying by 4–7 mm (example in Fig. 3c). Amoxicillin-clavulanic acid discs from Abtek showed acceptable variation with the susceptible QC strain but very large variation (9 mm, Fig. 3d) with the β-lactamase-producing QC strain, indicating that the content of clavulanic acid varied greatly from one disc to the other.

Discussion

Disc diffusion is an excellent, inexpensive and flexible method for AST but unless the quality of discs and media is tightly

Table 3
Variation in zone diameters (mm) for three agents from nine manufacturers when testing 20 consecutive disks from one vial. For the combination disks QC strains were chosen to challenge both the active agent and the inhibitor.

Antimicrobial disc and QC strain	Test results	Abtek	BD	Bio-analyse	Bio-Rad	HiMedia	Liofil-chem	Mast	Oxoid	SirScan	QC target (QC range)
Amoxicillin-clavulanic acid 20–10 μg versus *E. coli* ATCC 25922	Mean	19	19	19	22	19	17	21	20	19	21 (18–24)
	Min–Max	18–20	19–20	18–21	21–22	17–21	16–17	20–22	20–21	18–20	
	SD	0,7	0,5	0,9	0,2	1,1	0,5	0,6	0,5	0,6	
Amoxicillin-clav. 20–10 μg versus *E. coli* ATCC 35218	Mean	14	18	15	20	14	15	19	18	17	19–20 (17–22)
	Min–Max	8–16	18	12–17	19–20	11–17	15–16	18–19	18–19	16–17	
	SD	1,9	0,0	1,5	0,4	1,7	0,4	0,5	0,5	0,5	
Piperacillin-tazo. 30–6 μg versus *E. coli* ATCC 25922	Mean	21	23	24	23	22	22	25	23	24	24 (21–27)
	Min–Max	20–22	22–23	23–25	22–24	20–23	21–23	25–26	23–24	23–25	
	SD	0,4	0,3	0,6	0,6	0,9	0,6	0,5	0,3	0,7	
Piperacillin-tazobactam 30–6 μg versus *K. pneumoniae* ATCC 700603	Mean	14	17	17	17	15	16	19	17	18	17 (14–20)
	Min–Max	13–15	17–18	16–18	16–17	13–16	15–16	18–20	17	17–19	
	SD	0,6	0,4	0,4	0,5	1,0	0,3	0,5	0,0	0,6	
Tobramycin 10 μg versus *E. coli* ATCC 25922	Mean	21	24	23	23	28	23	23	24	23	22 (18–26)
	Min–Max	20–22	23–24	22–24	22–23	27–28	22–23	23–24	23–24	23–24	
	SD	0,6	0,4	0,6	0,3	0,5	0,4	0,4	0,3	0,2	

Values in shaded cells indicate variation >3 mm or standard deviation ≥1 mm for 20 disks from one vial.

controlled, variation may reach unacceptable levels. The EUCAST Development Laboratory investigated the activity and quality of 16 antimicrobial discs from nine manufacturers available on the international market in 2014 and 2017. Our results serve as a warning to manufacturers and users, but are also encouraging because a quality improvement was achieved where quality could be improved.

The EUCAST Development Laboratory does not have the resources to systematically evaluate commercial AST materials but will, on occasion, warn against products of doubtful quality [14–17]. The study performed in 2014 demonstrated large variation between nominally equipotent discs. The quality of discs from some manufacturers was unacceptable and many discs did not comply with the QC target values in EUCAST QC Tables. Some discs were clearly overcharged, which would appear as false susceptibility (very major error) in susceptibility test reports. Following the first study, EUCAST encouraged manufacturers to improve quality for discs with poor results and to withdraw disc lots with out-of-range results (categorized as red in this study). EUCAST also released a warning to clinical laboratories on the EUCAST website (www.eucast.org/warnings) against the use of disc lots with poor quality (categorized as red and orange in this study).

Results from the second study showed an overall improvement in disc quality and six of the nine manufacturers exhibited 100% of readings within QC ranges. Also, the number of discs giving results close to the target values clearly increased compared with the first study. HiMedia improved overall results from 67% to 83% of readings within QC ranges, but the performance was still unacceptable. Following the second study, manufacturers were again provided with support to improve the quality further. For example, Liofilchem has reviewed and adjusted their production of pefloxacin and amoxicillin-clavulanic acid discs and a small trial at the EUCAST Development Laboratory confirmed that the new lots were within acceptable limits.

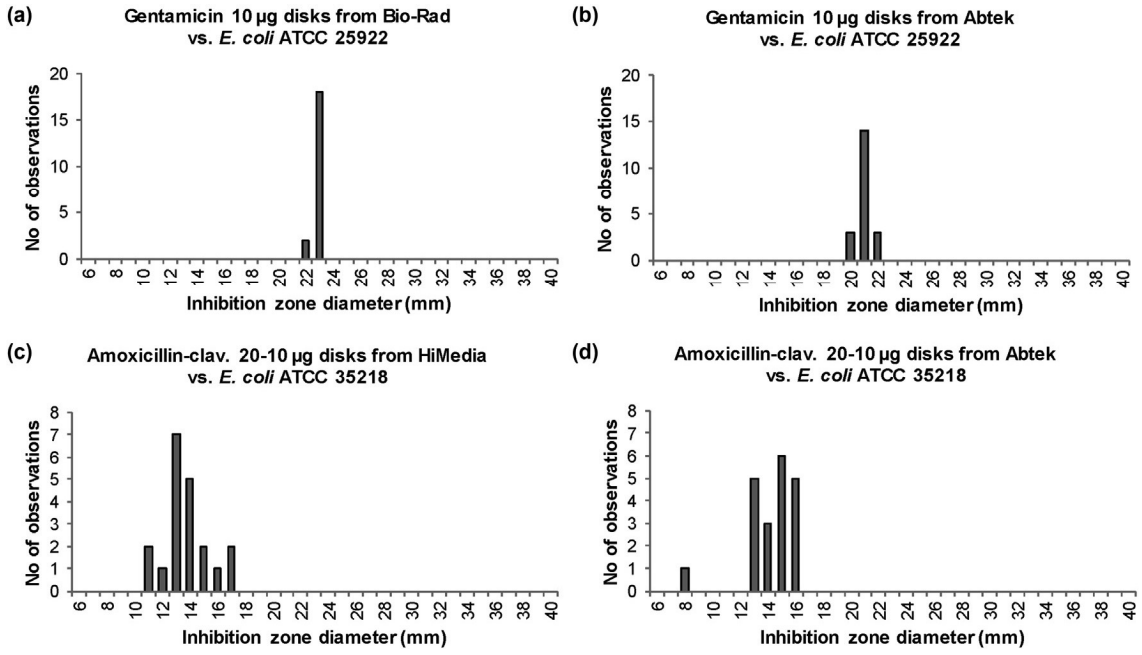

Fig. 3. Examples of disc-to-disc variation for 20 discs from one vial, showing good results (a, b) and unacceptable results (c, d).

Identifying unacceptable variation in activity between discs in a single vial adds a second dimension to the issue of disc quality. To our knowledge there are currently no official criteria for variation between discs within one vessel (vial, bottle, cartridge), but, from previous experience, we expected that ≥95% of discs would exhibit zone diameters distributed over a maximum of 2 mm, and that no disc would produce a zone outside the mean ± 1 mm. A variation of several millimetres, as found in several instances, is unacceptable. Equally serious, several manufacturers exhibited problems related to the inhibitor component of β-lactam–β-lactamase inhibitor combination discs. It is clear that for such discs, the QC must be directed both towards the active agent and the inhibitor.

EUCAST and CLSI [13,18] have published zone diameter QC criteria for several non-fastidious and fastidious QC strains for many years. The two committees agree on the QC ranges for non-fastidious QC organisms, when the disc contents are identical. In addition to ranges, EUCAST also lists target values, of importance to both manufacturers and users. For the laboratory, repeat testing of a QC strain should result in mean values close to the target values, whereas a random day-to-day variation inside the range is permitted. For manufacturers, the aim should be to produce discs resulting in zone diameters on or close to the target values for relevant QC strains. Hence, the range sets a standard for unavoidable random variation, whereas the target controls systematic errors. Even though manufacturers carry the main responsibility for the quality of the discs, the laboratory must ensure that discs are stored and handled as prescribed by the manufacturer [19]. This way, manufacturers and users have a shared responsibility for the quality of products used in AST. The final accuracy and precision of the method, as measured by laboratories through their internal QC procedures, is a product of the quality of the materials used and of the procedures as adopted and performed by the laboratory.

In this study, testing was performed under stringent and standardized conditions and discs were stored and handled strictly according to the manufacturers' instructions. Results were consistent and conclusive. Several discs contained amounts of active agent far above or below the nominal amount. This could be due to inaccurate production control, poor stability of the agent or a conscious over- or under-charging of the discs. The quality criteria set up by EUCAST are obviously possible to achieve as several manufacturers could, and others managed once the inadequacy of their discs was pointed out.

It is not uncommon for manufacturers to publish product-specific QC criteria that disagree with EUCAST criteria. For laboratories using EUCAST criteria, this is unacceptable. Furthermore, manufacturers sometimes claim that their products (discs, gradient tests) should only be used with their respective MH medium. In our opinion, this is only valid if manufacturers refuse to sell the products separately. EUCAST clinical breakpoints and QC criteria were developed using discs and media from several manufacturers so the system is set up to allow for variation caused by minor differences between materials from different manufacturers. Clinical laboratories should be able to combine MH agar from one manufacturer with antimicrobial discs from another, and still obtain reproducible and correct results.

Our study was limited to testing 16 selected antimicrobial discs, as testing of all available discs from nine manufacturers was not possible. A similar study should be conducted to evaluate the performance of MH agar from different manufacturers. The current study was performed using MH media from two major manufacturers and we have good experience from several more, but have not performed or seen a systematic comparison in modern times.

We encourage laboratories to check the performance of discs and immediately report problems to the manufacturer, and when unresolved, also to the EUCAST Development Laboratory. The manufacturer should take immediate action to resolve the problem. Major discrepancies, such as those observed in several discs in this study, are easily detected. If undetected, they may have dire consequences for the patient as the susceptibility test category may be susceptible instead of resistant or vice versa. Reporting organisms as false susceptible may jeopardize the life of the patient, while reporting organisms as false resistant may prevent efficient and safe therapy for the patient.

Conclusions

This evaluation points to excellent, acceptable and in some cases unacceptable quality of antimicrobial discs from nine manufacturers in two studies performed in 2014 and 2017. It shows that it is possible to produce discs of a high standard that repeatedly meet the QC targets, because at least three manufacturers showed excellent and consistent disc quality over a period of 3 years. It also demonstrates a general improvement of the quality of discs, where an improvement was possible, when the problems were pointed out to manufacturers and users. Our study points to the need for more stringent criteria for disc production and quality control. The current standards from the US Food and Drug Administration, Deutsches Institut für Normung and the World Health Organization provide criteria with very wide margins and only regulate the nominal amount of agent added to the disc, but none of them addresses the factual activity of the disc, which we consider more important than the amount added. These regulating documents, from an era when the quality of the paper used for antimicrobial discs was much poorer than today and when the know-how governing production had not reached today's status, are in need of more than a nominal update. Our study also points to the need for external evaluation of products used for AST.

Transparency declaration

GK has previously consulted for Oxoid Ltd on technical matters, but not after 2016. The other authors have no conflicts of interest to declare.

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Part of the results from this study have been presented as a poster at the 25th ECCMID, Copenhagen, Denmark, 2015 (Åhman J, Bengtsson S, Matuschek E and Kahlmeter G, Antimicrobial discs for susceptibility testing from nine manufacturers—uneven quality discovered. P 1239) and as a poster at the 28th ECCMID, Madrid, Spain, 2018 (Åhman J, Matuschek E, Varga Á and Kahlmeter G, Improved quality of antimicrobial discs for susceptibility testing from nine manufacturers. P 0156).

The study was not sponsored by the manufacturers of discs or media. All manufacturers were offered the same opportunity to discuss results of both studies (2014, 2017). A workshop for manufacturers, free of charge, was held in Copenhagen 6 November 2017, where all companies were invited and most were represented. Results and means for quality improvement were discussed openly.

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Author contribution

JÅ performed the antimicrobial susceptibility testing. JÅ, EM and GK have planned the study and analysed and evaluated the results. All of the authors have contributed to writing the manuscript.

Appendix A. Supplementary data

Supplementary data related to this article can be found at https://doi.org/10.1016/j.cmi.2018.05.021.

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