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Dear Colleague,

C.D.S. USERS' GROUP

Please find enclosed a questionnaire in regard to the planned workshop at A.S.M. We would be grateful if you would complete this and if you would like to discuss any aspect in more detail include your name and telephone number. Last year's workshop was so very well attended we were embarrassed by the restricted accommodation. This year, your early indication of attendance will help to avoid this problem and your advice will also assist us in the preparation of material for the workshop.

Also enclosed is Newsletter No. 4 which includes a major reformat of data relevant to the C.D.S. method. Your comments and criticisms of this change would be appreciated and if you wish you could include these with the completed questionnaire. Otherwise, if you have any comments, suggestions or questions you could telephone us on (02) 399-4053.

Finally, my attention has been drawn to problems some members of the group have been experiencing with prepared plates of Sensitest Agar from one particular supplier. On some batches of these plates the reference strain of Staphylococcus aureus NCTC 6571 grew poorly, if at all. The problem was investigated by the staff of the Antibiotics Laboratory in our department and found it to be due to the addition of sodium hydroxide to the medium during its preparation. The supplier reported that after ceasing this practice the most recent batches prepared on March 10, 1992, performed satisfactorily. The incident highlights one of the important quality assurance practices emphasized in the C.D.S. method, that is, to follow the manufacturer's directions precisely in procedures such as the reconstitution of dehydrated media. Media manufacturers should supply manuals free to all laboratories who request them and it is strongly suggested that those laboratories who do not have an "Oxoid Manual", for example, should obtain one.

We shall keep you informed of the venue, date and time of the Sydney workshop and I look forward to seeing you there.

Kind regards.

Yours faithfully,

S. M. BELL,
Department of Microbiology.

NO. 4

CDS USERS GROUP NEWSLETTER

Change in presentation of data:
In response to requests from users of the C.D.S. method we have extended the range of antibiotics calibrated and added a number of bacterial species which can be tested by the C.D.S. method. Because of the volume of the data so collected, they are more clearly presented in three separate tables:-

Table 1. Established calibrations.

This table lists the disc potencies and M.I.C. of susceptible strains for the more commonly encountered pathogens, all of which are tested in air at 35^0C.

Table 2. Provisional calibrations.

This table includes calibration results with organisms with unusual growth requirements and/or those pathogens uncommonly isolated in the routine laboratory. Either the rarity of these organisms or an absence in variation in the antibiotic susceptibility of the isolates encountered so far precludes an extensive comparison of zone sizes and M.I.C. For this reason these strains should be regarded as "provisionally calibrated" for the method and only further experience will confirm these calibrations.

Table 3. Reference strains.

This table lists the acceptable range of annular radii with each antibiotic when tested against the six reference strains currently used in the test for quality assurance purposes.

NOTE: The inclusion of an antibiotic with a particular reference strain in the table **does not** indicate that this antibiotic is tested in practice with that species. The inclusion of an antibiotic with one of the six reference strains is for quality assurance purposes only. The appropriate antibiotics to be tested and reported for each species are shown in Tables 1 and 2.

Increase of trimethoprim disc potency to 5 µg:
The potency of the trimethoprim disc was increased from 2.5 to 5.0 µg to overcome the interference with the test most probably arising from what would otherwise be regarded as minor variations in the constituents of Sensitest Agar. These variations have become evident over time and have resulted in an observed reduction in zone sizes with the reference strain of *E. coli* NCTC 10418.

Recently calibrated C.D.S. tests

A number of antibiotics and/or further species have been added to the range already tested by the C.D.S. method. Details of the changes are set out below:

i. Haemophilus influenzae

Two additional antibiotics have been calibrated for this species.

Oxytetracycline 30 µg. Oxytetracycline 30 µg discs yielded a much clearer distinction between resistant and susceptible strains of *H. influenzae* than did the tetracycline hydrochloride discs used with other species. Oxytetracycline 30 µg, therefore, is the disc which is to be used in testing *H. influenzae* and indicates susceptibility to other tetracyclines, including doxycycline.

Ciprofloxacin 2.5 µg. All strains tested so far were susceptible.

Cefaclor 30 µg. This disc was previously added to the range for *H. influenzae*. The results with this disc also can be used to indicate susceptibility to Augmentin.

NOTE: These three antibiotics, tetracycline, ciprofloxacin and cefaclor are intended for use with respiratory isolates of *H. influenzae* **ONLY**. It is not proposed that they be used for isolates from other sites or for type b strains.

ii. Listeria monocytogenes

For those who wish to test this organism two antibiotics have been calibrated on blood Sensitest Agar at 35^0C in air. All strains that we have tested so far were susceptible to ampicillin and gentamicin. The disc potencies used were ampicillin 25 µg and gentamicin 10 µg.

NOTE: Ampicillin and **NOT** benzylpenicillin is used to test penicillin susceptibility of this species.

iii. Neisseria meningitidis

The medium for testing this species is blood Sensitest Agar incubated at 35^0C in **5% carbon dioxide**. Three antibiotics, benzylpenicillin, cefotaxime and chloramphenicol, have been calibrated and it was found that all isolates tested so far were susceptible to these antibiotics.

iv. Staphylococcus saprophyticus

This species of coagulase-negative staphylococcus is most simply defined as a coagulase-negative staphylococcus resistant to novobiocin 5 µg and may cause urinary tract infections in domiciliary patients.

Amoxycillin 10 µg. This antibiotic was found to be the most useful agent to indicate susceptibility to benzylpenicillin, ampicillin and amoxycillin.

NOTE: Amoxycillin 10 µg is restricted for use with *Staphylococcus saprophyticus* isolated from urinary tract infections only. **It should not be used for testing any other staphylococcus species.**

v. Branhamella catarrhalis

Five antibiotics have been calibrated with this species and are tested on blood Sensitest Agar at 35^0C in **5% carbon dioxide**. The antibiotics calibrated are benzylpenicillin, cefaclor, ciprofloxacin, erythromycin and tetracycline. As with *H. influenzae*, susceptibility to cefaclor also indicates susceptibility to Augmentin.

Table 1. Established calibrations: Disc potencies and MIC of susceptible strains for common pathogens, all tested in air, at 35°C using Sensitest Agar unless indicated.

Antibiotic	Disc potency (µg)		MIC for susceptible strains (mg/L)
Staphylococci, streptococci[1] and pneumococci[1]			
Benzylpenicillin	0.5 u		≤ 0.06
Chloramphenicol	30		≤ 8.0
Ciprofloxacin	*(Staph. only)* 2.5		≤ 1.0
Erythromycin	5		≤ 0.5
Fusidic acid	2.5		≤ 0.5
Gentamicin	10		≤ 1.0
Kanamycin	50		≤ 4.0
Methicillin	5		≤ 4.0
Rifampicin	1		≤ 0.5
Tetracycline	30		≤ 2.0
Vancomycin	60	4 mm[3]	≤ 2.0
Enterobacteriaceae			
Amikacin	30		≤ 4.0
Ampicillin	25		≤ 8.0
Augmentin	*(Urine only)*	60	≤ 8.0/4.0
Aztreonam	10		≤ 4.0
Cefotaxime	5		≤ 1.0
Cefotetan	10		≤ 4.0
Cefoxitin	30		≤ 8.0
Ceftazidime	10		≤ 4.0
Ceftriaxone	5		≤ 2.0
Cephalexin	*(Urine only)*	100	≤ 16.0
Chloramphenicol	30		≤ 8.0
Ciprofloxacin	2.5		≤ 1.0
Gentamicin	10		≤ 1.0
Imipenem	10		≤ 4.0
Kanamycin	50		≤ 8.0
Nalidixic acid	*(Urine only)*	30	≤ 4.0
Netilmicin	30		≤ 2.0
Nitrofurantoin	*(Urine only)*	200	≤ 32.0
Norfloxacillin	*(Urine only)*	10	≤ 4.0
Sulphafurazole	300		≤ 64.0
Tetracycline	30		≤ 4.0
Timentin	85		≤ 32.0/2.0
Tobramycin	10		≤ 1.0
Trimethoprim	5		≤ 2.0
Pseudomonas aeruginosa			
Amikacin	30	4 mm[3]	≤ 16.0
Aztreonam	30		≤ 8.0
Ceftazidime	10		≤ 4.0
Ciprofloxacin	2.5		≤ 2.0
Gentamicin	10	4 mm[3]	≤ 4.0
Imipenem	10		≤ 4.0
Netilmicin	30	4 mm[3]	≤ 8.0
Piperacillin	50		≤ 16.0
Polymyxin	300 u	4 mm[3]	≤ 1.0
Ticarcillin	75		≤ 32.0
Timentin	85		≤ 32.0/2.0
Tobramycin	10	4 mm[3]	≤ 4.0
Haemophilus influenzae[2]			
Ampicillin	2		≤ 1.0
Cefaclor	*(Sputum only)*	30	≤ 4.0
Cefotaxime	5		≤ 0.12
Chloramphenicol	10		≤ 2.0
Ciprofloxacin	*(Sputum only)*	2.5	≤ 1.0
Oxytetracycline	*(Sputum only)*	30	≤ 4.0

[1] Test on blood Sensitest Agar.

[2] Test on chocolated blood agar (Columbia Agar Base).

[3] The annular radius of the zone of inhibition for sensitive strains is ≥ 4 mm.

Table 2a. Provisional calibrations: Disc potencies, MIC of susceptible strains, media used and incubation conditions for uncommon pathogens.

Antibiotic	Disc potency (µg)		MIC for susceptible strains (mg/L)
***Acinetobacter* species**			
(Sensitest, air, 35°C)			
Amikacin	30		≤ 4.0
Ampicillin	25		≤ 8.0
Ciprofloxacin	2.5		≤ 1.0
Gentamicin	10		≤ 1.0
Imipenem	10		≤ 4.0
Netilmicin	30		≤ 2.0
Norfloxacillin	10		≤ 4.0
Sulphafurazole	300		≤ 64.0
Ticarcillin	75		≤ 32
Tobramycin	10		≤ 1.0
Branhamella catarrhalis			
(Blood Sensitest, CO_2, 35°C)			
Benzylpenicillin	0.5 u		≤ 0.12
Cefaclor	30		≤ 4.0
Ciprofloxacin	*2.5*		≤ 1.0
Erythromycin	5		≤ 0.5
Tetracycline	30		≤ 4.0
Enterococci			
(Blood Sensitest, air, 35°C)			
Ampicillin	25		≤ 8.0
Gentamicin	200	4 mm**1**	≤ 512
Vancomycin	60	4 mm**1**	≤ 2.0
Listeria monocytogenes			
(Blood Sensitest, air, 35°C)			
Ampicillin	25		≤ 8.0
Gentamicin	10		≤ 1.0
Neisseria meningitidis			
(Blood Sensitest, CO_2, 35°C)			
Benzylpenicillin	0.5 u	4 mm**1**	≤ 0.12
Cefotaxime	5		≤ 0.12
Chloramphenicol	10		≤ 2.0

1 The annular radius of the zone of inhibition for sensitive strains is ≥ 4 mm.

Table 2b. Provisional calibrations continued: Disc potencies, MIC of susceptible strains, media used and incubation conditions for uncommon pathogens.

Antibiotic	Disc potency (µg)		MIC for Susceptible strains (mg/L)
Coagulase-negative staphylococci from URINE ONLY (Sensitest, air, 35°C)			
Amoxycillin *(Staph. sapro. only)*	10		≤ 0.5
Benzylpenicillin[2]	0.5 u		≤ 0.06
Chloramphenicol	30		≤ 8.0
Ciprofloxacin	2.5		≤ 1.0
Erythromycin	5		≤ 0.5
Fusidic acid	2.5		≤ 0.5
Gentamicin	10		≤ 1.0
Kanamycin	50		≤ 4.0
Methicillin[2]	5		≤ 4.0
Nitrofurantoin	200		≤ 32.0
Rifampicin	1		≤ 0.5
Sulphafurazole	300		≤ 64.0
Tetracycline	30		≤ 2.0
Vancomycin	60	4 mm[1]	≤ 2.0
Xanthomonas maltophilia (Sensitest, air, 35°C)			
Sulphafurazole	300		≤ 64.0
Yersinia enterocolitica (Sensitest, air, 30°C)			
Amikacin	30		≤ 4.0
Augmentin	3		≤ 2.0/1.0
Aztreonam	10		≤ 4.0
Chloramphenicol	30		≤ 8.0
Ciprofloxacin	2.5		≤ 1.0
Gentamicin	10		≤ 1.0
Imipenem	10		≤ 4.0
Netilmicin	30		≤ 2.0
Sulphafurazole	300		≤ 64.0
Tetracycline	30		≤ 4.0
Timentin	85		≤ 32.0/2.0
Tobramycin	10		≤ 1.0
Trimethoprim	5		≤ 2.0

1. The annular radius of the zone of inhibition for sensitive strains is ≥ 4 mm.
2. Benzylpenicillin and methicillin are not used for testing *S. saprophiticus.*

Table 3a. Reference strains: Antibiotic disc content and the acceptable range (mm) of the annular radii of inhibition with the reference strains used in the CDS method.

Antbiotic	Disc content (µg)	Acceptable range* (mm)
Staphylococcus aureus **NCTC 6571**		
Amoxycillin	10	11.5 - 15.9
Benzylpenicillin	0.5 u	8.7 - 13.5
Chloramphenicol	30	7.8 - 11.4
Ciprofloxacin	2.5	9.2 - 12.4
Erythromycin	5	7.1 - 10.7
Fusidic acid	2.5	8.6 - 12.6
Gentamicin	10	6.6 - 9.4
Kanamycin	50	5.9 - 8.7
Methicillin	5	8.8 - 12.0
Nitrofurantoin	200	6.7 - 10.3
Rifampicin	1	9.3 - 12.5
Sulphafurazole	300	9.3 - 13.7
Tetracycline	30	10.6 - 16.2
Trimethoprim	5	7.3 - 10.1
Vancomycin	60	5.4 - 7.8
Haemophilus influenzae **NCTC 4560**		
Ampicillin	2	6.0 - 9.2
Chloramphenicol	10	7.7 - 10.9
Cefaclor	30	7.3 - 10.9
Cefotaxime	5	8.9 - 14.1
Ciprofloxacin	2.5	9.7 - 14.9
Oxytetracycline	30	6.6 - 9.0
Yersinia enterocolitica **IP 22273**		
Amikacin	30	6.0 - 8.4
Aztreonam	10	10.1 - 13.7
Augmentin	3	6.4 - 8.4
Chloramphenicol	30	6.7 - 11.9
Ciprofloxacin	2.5	12.1 - 16.1
Gentamicin	10	6.0 - 8.0
Imipenem	10	11.1 - 15.1
Netilmicin	30	8.1 - 10.5
Sulphafurazole	300	8.9 - 13.1
Tetracycline	30	9.9 - 13.9
Timentin	85	10.6 - 15.4
Tobramycin	10	6.1 - 8.1
Trimethoprim	5	9.6 - 13.2

* The acceptable range (95% confidence limits) is the mean ± 2 standard deviations.

Table 3b. Reference strains (continued): Antibiotic disc content and the acceptable range (mm) of the annular radii of inhibition with the reference strains used in the CDS method.

Antbiotic	Disc content (µg)	Acceptable range* (mm)
Escherichia coli **NCTC 10418**		
Amikacin	30	6.7 - 10.3
Ampicillin	25	7.5 - 10.7
Aztreonam	10	11.8 - 14.2
Cefotaxime	5	9.7 - 13.7
Cefotetan	10	11.6 - 13.6
Cefoxitin	30	9.8 - 13.0
Ceftazidime	10	8.7 - 11.9
Ceftriaxone	5	10.5 - 14.3
Cephalexin	100	6.9 - 10.9
Chloramphenicol	30	8.7 - 11.9
Ciprofloxacin	2.5	12.4 - 15.8
Gentamicin	10	6.2 - 9.4
Imipenem	10	10.3 - 13.5
Kanamycin	50	6.2 - 11.8
Nalidixic acid	30	8.9 - 12.1
Netilmicin	30	7.7 - 11.3
Nitrofurantoin	200	6.3 - 9.5
Norfloxacin	10	10.4 - 16.4
Sulphafurazole	300	5.0 - 9.4
Tetracycline	30	5.8 - 11.0
Tobramycin	10	6.4 - 8.4
Trimethoprim	5	8.7 - 11.1
Escherichia coli **NCTC 11560**		
Augmentin	60	6.4 - 9.6
Timentin	85	6.0 - 8.4
Pseudomonas aeruginosa **NCTC 10662**		
Amikacin	30	7.4 - 10.6
Aztreonam	30	8.3 - 13.1
Ceftazidime	10	7.5 - 11.9
Ciprofloxacin	2.5	8.9 - 14.5
Gentamicin	10	5.5 - 9.5
Imipenem	10	7.9 - 10.3
Netilmicin	30	6.4 - 10.4
Piperacillin	50	8.1 - 12.9
Polymyxin	300 u	5.2 - 7.2
Ticarcillin	75	7.3 - 12.1
Tobramycin	10	7.0 - 10.6

* The acceptable range (95% confidence limits) is the mean ± 2 standard deviations.