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European harmonization of MIC breakpoints for antimicrobial susceptibility testing of bacteria

Gunnar Kahlmeter[1]*, Derek F. J. Brown[2], Fred W. Goldstein[3], Alasdair P. MacGowan[4],
Johan W. Mouton[5], Anders Österlund[1], Arne Rodloff[6], Martin Steinbakk[7],
Pavla Urbaskova[8] and Alkiviadis Vatopoulos[9]

[1]*Klinisk mikrobiologi, Centrallasarettet, 351 85 Växjö, Sweden;* [2]*Clinical Microbiology and Public Health Laboratory, Addenbrooke's Hospital, Hills Road, Cambridge CB2 2QW;* [4]*Department of Medical Microbiology, Southmead Hospital, Westbury-on-Trym, Bristol BS10 5NB, UK;* [3]*Laborataire de Microbiologie Médicale, Hôpital Saint-Joseph, 185 rue Raymond Losserand, F-75014 Paris, France;* [5]*Department of Medical Microbiology and Infectious Diseases, Canisius Wilhelmina Hospital C-70, Weg door Jonkerbos 100, 6532 sz Nijmegen, The Netherlands;* [6]*Institut für Medizinische Mikrobio der Universitat Leipzig, Liebigstr 24, Leipzig 04103, Germany;* [7]*Department of Microbiology, Akershus University Hospital, P.O. Box 23, N-1474 Nordbyhagen, Norway;* [8]*National Reference Laboratory for Antibiotics, National Institute of Public Health, Srobarova 48, 100 42 Prague 10, Czech Republic;* [9]*Dept of Hygiene & Epidemiology, Medial School, Athens University, 75 M.Asias Str, GR-11527 Athens (Goudi), Greece*

The success or failure of antimicrobial therapy in bacterial and fungal infections is predicted ideally by antimicrobial susceptibility testing (AST), in which microorganisms are divided into treatable and non-treatable categories on the basis of MIC breakpoints. In Europe, the categorization was traditionally a clinical one and it was made irrespective of whether or not the organism harboured resistance mechanisms. MIC breakpoints generally divide bacteria into three categories of susceptibility: susceptible, intermediate or indeterminate, or resistant. These terms can be defined as susceptible (S—where the antimicrobial activity is associated with a likelihood of therapeutic success), intermediate or indeterminate (I—where the antimicrobial activity is associated with an indeterminate or uncertain therapeutic effect) and resistant (R—where the antimicrobial activity is associated with a higher than expected likelihood of therapeutic failure). MIC breakpoints are used either directly, as in MIC determination and 'breakpoint' susceptibility testing methods in broth or agar, or indirectly when converted into inhibition zone diameters in disc diffusion techniques.

The last decade has demonstrated that breakpoints need a new function: to detect the biological phenomenon of phenotypical resistance, and to monitor the development of antimicrobial resistance. The need for this new function has shown us firstly, that breakpoints designed to guide therapy do not necessarily distinguish between bacteria with and without resistance mechanisms, and do not necessarily allow their early and/or reproducible detection. Secondly, it has demonstrated that the lack of harmonized breakpoints among

methods in different countries, or even within the same country, often obviates meaningful comparison of resistance rates, monitoring of development of resistance in international surveillance systems and investigation of the effects of intervention strategies. Breakpoints have evolved to try to satisfy both the need to guide therapy and the need to detect biological resistance, often resulting in compromises that satisfy neither.

MIC breakpoints may be defined by national breakpoint committees and/or regulatory authorities, including medicines' agencies, such as the European Medicines Evaluation Agency (EMEA) in Europe, and the Food and Drug Administration (FDA) in the USA. In Europe, there are several active national breakpoint committees, including CA-SFM[1] in France, the DIN[2] in Germany, the CRG[3] in the Netherlands, the NWGA[4] in Norway, the SRGA[5] in Sweden and the BSAC Working Party on Antimicrobial Sensitivity Testing[6] in the UK. Each committee is long established, has 8–15 members from the fields of clinical microbiology, infectious diseases, pharmacology and, in some cases, from other fields of medicine. Each committee has devised a process for defining breakpoints. Several also have described AST methodologies to go with their breakpoints, but with few exceptions these methodologies are limited to their respective country. The equivalent committee in the USA is the NCCLS[7] with many adherents worldwide, including European countries.

It seems little more than chance that the committees occasionally recommend the same breakpoints and use the same terminology and/or methods. For example, the quite remarkable differences in cefo-

*Corresponding author. Tel: +46-470587477; Fax: +46-470587455; E-mail: gunnar.kahlmeter@ltkronoberg.se

Table 1. Similarities and differences in international breakpoint systems—current cefotaxime and ciprofloxacin breakpoints for Enterobacteriaceae in Europe and the USA

Breakpoint committee (country)	Cefotaxime breakpoint (mg/L)		Ciprofloxacin breakpoint (mg/L)	
	susceptible	resistant	susceptible	resistant
BSAC (UK)	≤2	≥4	≤1	≥2
CA-SFM (France)	≤4	>32	≤1	>2
CRG (Netherlands)	≤4	>16	≤1	>2
DIN (Germany)	≤2	≥16	≤1	≥4
NCCLS (USA)	≤8	≥64	≤1	≥4
NWGA (Norway	≤1	≥32	≤0.12	≥4
SRGA (Sweden)	≤0.5	≥2	≤0.12	≥2

taxime and ciprofloxacin breakpoints for Enterobacteriaceae are shown in Table 1. The difference in terminology is exemplified by the different signs used for separating the intermediate and resistant categories, CA-SFM and CRG using R > X mg/L, whereas the others, including the NCCLS, use R ≥ X mg/L. Although MIC determination methodologies are similar, all having the same origin, there are some differences in the recommended media. For disc diffusion methods the differences are even greater. European committees normally recommend a lighter inoculum than the confluent inoculum recommended by NCCLS, and there are differences in media, disc contents, incubation conditions and other technical details. The lack of harmonization of breakpoints, terminology and methods creates difficulties in the comparison of results of antimicrobial surveillance, and in communication between the medical profession, the pharmaceutical industry and the regulatory authorities. Antimicrobial chemotherapy is generally similar in different countries and it is not logical for the same organism to be regarded as susceptible in some countries whereas resistant in others.

MIC breakpoints are defined against a background of data, including intended or approved indications, clinical response data, dosing schedules, pharmacokinetics and pharmacodynamics, and microbiological data in a variety of formats. The process of setting breakpoints never was, and probably never will be, exact or strictly scientific; it contains elements of philosophy, 'fairness' and compromise. One might be sceptical at the introduction of a 'shift factor' as part of formulae otherwise containing fairly exact measurements, but without a shift factor to take account of microbiological distribution of susceptibility we arrive at systems lacking in practicality and reproducibility. However, whereas the 'shift' can be explained, it cannot totally be controlled.

In 1997, the European Society of Clinical Microbiology and Infectious Diseases (ESCMID) formed the European Committee on Antimicrobial Susceptibility Testing (EUCAST) with the intention that it act as a European breakpoint committee comparable with the NCCLS. Appropriate professional bodies from Europe, the pharmaceutical industry and AST device and media manufacturers have named representatives on the EUCAST. A number of subcommittees were set up to cover various aspects of susceptibility testing, including terminology, breakpoints and methodology, and several guidelines have been produced.[8–15] However, the EUCAST had no formal relationship with the European national breakpoint committees, and

there was no collaboration or coordination among the national committees. Harmonization remained elusive.

In 2001/2002 the ESCMID reorganized the EUCAST, with national committees being given a greater role. A chairman, a scientific secretary, representatives of active national breakpoint committees (presently the committees in France, Germany, Norway, Sweden, the Netherlands and the UK) and two representatives of the EUCAST General Committee (at the moment from the Czech Republic and Greece) formed the EUCAST Steering Committee. The General Committee has representatives from almost all European countries, and the pharmaceutical and device industry. The organization is detailed on the EUCAST website (www.eucast.org). A decision-making process, involving consultation with the EUCAST General Committee, but leaving the final decisions in the hands of the Steering Committee, was established. The major tasks of the EUCAST are to harmonize MIC breakpoints across Europe and to agree on a common reference methodology. An agar dilution technique has already been described,[10] and a broth microdilution technique will soon be published.[12] These methods are relatively standardized worldwide, and the EUCAST techniques are very similar to the corresponding techniques described by European national committees and the NCCLS, against which most commercial AST devices are calibrated. The EUCAST will liaise with other groups on initiatives in antimicrobial surveillance, and with regulatory agencies, the NCCLS, the drug industry, and manufacturers of discs, media and devices on relevant aspects of antimicrobial susceptibility testing.

In an attempt to overcome the problems of national differences in breakpoints based on clinical or microbiological data, the EUCAST will define separate breakpoints for, on the one hand, the detection of bacteria with resistance mechanisms and the monitoring of resistance development (epidemiological cut-off values) and, on the other, the guidance of therapy (clinical breakpoints). In order to achieve this the EUCAST will define the wild-type (WT) distributions of bacteria and (together with the EUCAST subcommittee on antifungal susceptibility testing) fungi, for relevant drug–bug combinations, that is, populations of organisms with no acquired phenotypically detectable resistance mechanism. The EUCAST is in the process of collecting full range MIC data from as many sources as possible (breakpoint committees, antimicrobial surveillance systems in man and animals, pharmaceutical companies, scientific reports, etc.). The data are entered into a database, each distribution is screened for acceptance, and then made available free to all on the Internet. For each combination of drug and species, all accepted MIC data are aggregated, together forming an international reference database of wild-type MIC distributions. As an example, the aggregated wild-type MIC distribution for *Escherichia coli* against ciprofloxacin is shown in Figure 1. Having determined the wild-type distribution and its highest MIC, organisms with acquired resistance mechanisms can be identified readily as organisms with reduced susceptibility compared with the highest MIC value of the wild-type. The EUCAST has defined this MIC value tentatively as the 'epidemiological cut-off value' or wild-type (WT) cut-off value. The term 'microbiological breakpoint' has not been used deliberately to avoid confusion with the current understanding of 'breakpoint' as indicating a clinical breakpoint. The epidemiological cut-off value is expressed as WT ≤ X mg/L (in Figure 1 it is defined as WT ≤ 0.064 mg/L). In analogy with resistance, non-WT is defined as >X mg/L.

Wild-type MIC distributions are needed to define epidemiological cut-off values, and to ensure that clinical breakpoints do not divide
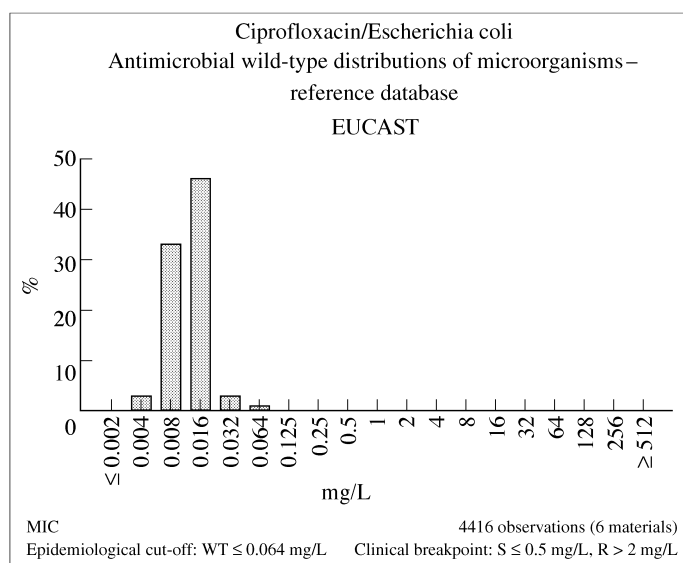
Ciprofloxacin/Escherichia coli
Antimicrobial wild-type distributions of microorganisms –
reference database
EUCAST

MIC   4416 observations (6 materials)
Epidemiological cut-off: WT ≤ 0.064 mg/L   Clinical breakpoint: S ≤ 0.5 mg/L, R > 2 mg/L

Figure 1. The EUCAST reference wild-type distribution for *E. coli* tested against ciprofloxacin (from http://www.srga.org/eucastwt/WT_EUCAST.htm). Aggregated distribution of data from six sources (4416 observations) from breakpoint committees, antimicrobial surveillance systems, drug companies and scientific publications. The EUCAST tentative 'epidemiological cut-off value' (WT ≤ 0.064 mg/L) and harmonized tentative clinical breakpoints for Enterobacteriaceae (S ≤ 0.5 and R > 2 mg/L) are shown.

wild-type populations of important species. The EUCAST wild-type distribution will also provide a reference MIC distribution against which methods can be calibrated. Epidemiological cut-off values are needed to prevent the ongoing compromise between clinical and epidemiological aspects of the detection of resistance. They will be valuable especially (i) when the gap in MIC between the upper end of the wild-type distribution and the clinical breakpoint is great, (ii) when the clinical breakpoint divides wild-type distributions of bacteria (something which breakpoint committees should avoid at all costs) (iii) where clinical breakpoints or consensus on clinical breakpoints are lacking, in which case epidemiological cut-off values allow the detection and comparison of resistance, (iv) for new and old drugs where resistance has yet to be described (for example, *Streptococcus pyogenes* and penicillin), (v) in situations where it is important that 'resistance' represents microorganisms with resistance mechanisms (for example, in studies of the relationship between the use of antimicrobials and resistance, and studies investigating strategies to counteract resistance development, both of which are dependent on high sensitivity and high specificity).

Clinical breakpoints are the breakpoints used to guide therapy and are expressed as S ≤ X mg/L and R > Y mg/L. In the interests of a common terminology, the national breakpoint committee representatives on the EUCAST Steering Committee have agreed to express the resistant category as R > Y mg/L rather than R ≥ Y mg/L. This avoids a gap in the distribution that otherwise exists between two-fold dilutions. In Figure 1, the clinical breakpoints are defined as S ≤ 0.5 and R > 2 mg/L (tentative EUCAST clinical breakpoints for ciprofloxacin and Enterobacteriaceae), whereby strains with MICs > 0.5 and ≤ 2 mg/L are intermediate.

Agreement on the epidemiological cut-off values should not be difficult as the wild-type MIC distributions make these more or less self-evident. Harmonizing 'clinical breakpoints' is less of a scientific process, leaving room for interpretation and opinion. The lack of sci-

entific evidence of clinical effect often hampers the process. The use of pharmacokinetic/pharmacodynamic data and 'Monte Carlo simulation'[16] provide a basis for decisions on breakpoints, especially on how to define clinical susceptibility of bacteria belonging to the wild-type. The fact that we do not need to compromise between epidemiological and clinical aspects will facilitate the discussion. In addition, the process of harmonization of clinical breakpoints is assisted if it is accepted that they must not divide wild-type distributions of important or common species, as such a division leads to lack of reproducibility and is scientifically unsound when the wild-type population is homogeneous. We have applied this method to produce tentative fluoroquinolone breakpoints and wild-type cut-off values, and are currently addressing the aminoglycosides, glycopeptides and linezolid using the same approach. These recommendations will be available for consultation shortly. We expect that these ongoing programmes of work, once concluded, will allow the EUCAST, in concert with the European national breakpoint committees, to put in place a set of European breakpoints and epidemiological cut-off values. This should aid detection and monitoring of antibiotic resistance, and assist in new drug and device development in Europe and elsewhere.

Acknowledgements

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