Microbiology guide to interpreting minimum inhibitory concentration (MIC)

Historically, *in vitro* susceptibility testing was routinely performed by disk diffusion (Kirby-Bauer) method. The size of the growth-free zone determined whether the bacterium was considered to be susceptible, resistant, or intermediate to a particular antibiotic.

Although a useful guide for selecting an effective antibiotic, Kirby-Bauer testing could not tell the clinician the exact concentration of antibiotic needed to achieve a therapeutic result. The alternative automated quantitative method of susceptibility testing with the Vitek platform used in our microbiology laboratories supports rapid and accurate quantitative antibiotic susceptibility test (AST) reporting, including minimum inhibitory concentration (MIC). The MIC provides the ability to precisely determine the concentration of antibiotic required to inhibit growth of a pathogen.

Your IDEXX microbiology results will show the identity of the organism and the appropriate antibiotic sensitivity pattern against each organism. Most antibiograms will include MICs in order to determine the most effective antibiotic that will result in effective treatment.

This guide provides a detailed explanation of the following concepts which are important in implementing the MIC:

- The MIC number is the lowest concentration (in μg/mL) of an antibiotic that inhibits the growth of a given strain of bacteria. (See the section "What is an MIC?")
- An MIC number for one antibiotic CANNOT be compared to the MIC number for another antibiotic. (See the "How are MICs used?" section.)
- The choice of antibiotic should not be based on the MIC alone but on several other criteria. (See the "antibiotic selection" section).
- The attached tables will aid in MIC interpretation and antibiotic selection.

What is an MIC?

The MIC, or minimum inhibitory concentration, is the lowest concentration (in μg/mL) of an antibiotic that inhibits the growth of a given strain of bacteria. This is a strain dependent value that will greatly differ between different bacterial species and even within one species depending on the individual strain isolated in the respective clinical case, even for the same antibiotic examined. At IDEXX, a commercial automated system is used to determine MICs. A quantitative method of susceptibility testing, an MIC helps determine which class of antibiotic is most effective. This information can lead to an appropriate choice of an antibiotic that will increase chances of treatment success and help in the fight to slow antibiotic resistance.

The MIC is reported as a numerical value. Values above or below the range of measurement are indicated with "<=" (in case of below the range, in the susceptible category) or with ">=" (in case of above the range, in the resistant category).

How is the MIC reported?

Next to each antibiotic is the susceptibility interpretation: S (sensitive), I (intermediate), or R (resistant) are the three interpretative categories, listed in the first result column of the susceptibility report. The respective category is followed by the MIC in μg/mL in the next result column of the susceptibility report. "Sensitive" implies that the organism is inhibited by the serum concentration of the drug that is achieved using the usual dosage; "intermediate" implies that the organisms are inhibited only when higher concentrations than with the usually recommended dosages are achievable; and "resistant" implies that the organisms are resistant to the usually achievable serum drug levels. These interpretive standards have been established by the Clinical and Laboratory Standards Institute (CLSI). The interpretative categories are evaluated according to the so-called breakpoints for each antibiotic as listed in the current versions of the CLSI documents.

Example for reporting AST result

Bacteriologic examination, culture aerobic:
- *Staphylococcus intermedius* group, moderate count (1)
- *Escherichia coli*, high count (2)

Antibiogram for strain no.	(1)		(2)		Remark
	SIR	MIC	SIR	MIC	
Penicillin	R	>=0.5			
Ampicillin	R		S	<=2	Also valid for Amoxicillin
Amoxicillin	R		S		Also valid for Ampicillin
Amoxicillin/clavulanic acid	S	<=2	S	<=2	
Cefalexin	S		S	<=4	
Cefazolin	S				
Cefalotin	S	<=2	I	4	
Cefovecin	S	<=0.5	S	<=0,5	
Ceftiofur	S	<=0.5	S	<=1	
Cefpodoxim	S		S	<=0,25	
Cefoperazone	S		S		
Cefquinome	S		S		
Oxacillin	S	<=0.25	S		
Cloxacillin	S				
Gentamicin	S	<=0.5	S	<=1	
Kanamycin	S	<=4			Also valid for Neomycin
Erythromycin	S	0.5			
Tetracycline	S	<=1	S	<=1	Stands for all tetracyclines
Doxycycline	S	<=0.5	S	1	
Enrofloxacin	S	<=0.5	S	0,5	Stands for all fluorchinolones
Marbofloxacin	S	<=0.5	S	<=0,5	
Pradofloxacin	S	<=0.12	S	0,25	
Nitrofurantoin	S	<=16	S	<=16	
Clindamycin	S	0.25			
Lincomycin	S				
Sulfamethox./Trim.	S	<=10	S	<=20	Also valid for other sulfonamide/trim. combinations
Chloramphenicol	S	8	S	8	Stands for all phenicoles (e.g. florfenicol)
Florfenicol	S		S		
Imipenem	S		S	<=0,25	Strictly verify indication
Meropenem	S		S		Strictly verify indication
Amikacin	S		S	<=2	Strictly verify indication
Tobramycin	S		S		Strictly verify indication
Azithromycin	S				Strictly verify indication
Clarithromycin	S				Strictly verify indication

Interpretation key for the antibiogram:

S = sensitive; active substance normally effective against microorganisms at recommended dosage.
I = intermediate; active substance may be effective against microorganism at higher than recommended dose.
R = resistant; active substance not effective against microorganism either in recommended or higher dosage due to resistance mechanism.

Method: automated resistance determination (MIC), according to guidelines of the Clinical and Laboratory Standards Institute (CLSI). MIC values (if available) expressed in µg/ml.

SIR results and / or MIC values are not reported if no CLSI interpretation criteria (so-called breakpoints) are available, the growth conditions of the microorganism do not permit measurement with the method used, or determination for the individual isolate was not possible.

The results are partly obtained by derivation according to international guidelines (information available on request from the laboratory). The antibiogram is compiled according to the microorganisms. An absent SIR result in the antibiogram generally indicates an unsuitable combination of microorganism and active substance. Please also note the information regarding lead substances and cross resistances.

Please observe application restrictions and contraindications! The choice of antibiotic is the responsibility of the attending veterinarian. The test method refers to systemic effective levels. Testing locally applicable active substances such as fusidic acid and polymyxin B is not possible.

For more information about the MIC, please visit our website (keyword „MIC“).

When are MICs not performed?

MICs are not performed when:

- The growth requirements of some organisms require the sensitivity testing to be performed by another method.
- Interpretive criteria are not available from CLSI.
- Certain antibiotics are not available on our automated system.
- The drug is known to be clinically ineffective against the organism, regardless of the *in vitro* results.
- By design the drug is not active against the microorganism, e.g. Clindamycin is designed for Gram-positive microorganisms only, and therefore not tested against Gram-negative microorganisms (e.g. Pseudomonas sp.)
- Due to the antibiotic panel in the automated system the antibiotic was not measured, and the result for the interpretative category was achieved by deduction according to international guidelines.

How are MICs used?

An antibiotic breakpoint is the dilution where bacteria begin to show resistance. The breakpoint and range of dilutions differ by drug and bacterial species. Tables listing the respective breakpoints are subject to constant re-evaluation by CLSI according to newest scientific data (pharmacological and clinical data). The most current versions are available online (see **https://clsi.org/**).

Comparing MICs of different antibiotics is not based solely on the numerical value but on how far the MIC is from the breakpoint, the site of the infection, and other considerations, such as the age, species, and health of the animal. Possible side effects of the drug, frequency, and route of administration are also important factors.

For example: A strain of *Staphylococcus pseudintermedius* has an MIC of 0.5 µg/ml for erythromycin and of 0.25 µg/ml for pradofloxacin. Looking at the dilutions for erythromycin, at 0.5 µg/ml, this strain of *Staphylococcus pseudintermedius* is four dilutions away from the breakpoint. For pradofloxacin, the same strain of *Staphylococcus pseudintermedius* at an MIC of 0.25 µg/ml is three dilutions away from the breakpoint. So, based on MICs, this strain of *Staphylococcus pseudintermedius* is more susceptible to erythromycin than pradofloxacin. For other factors to take into consideration see section "antibiotic selection".

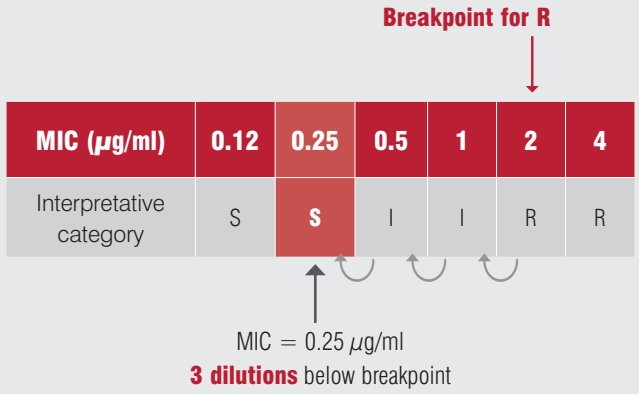
Quick guide for MIC interpretation

See below a simplified approach to use MIC values:

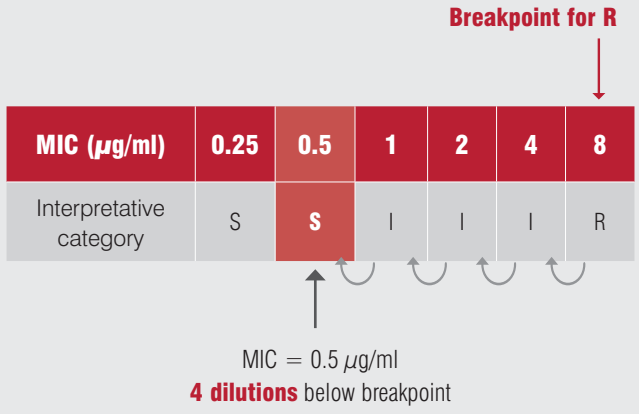
If the result is:	MIC	Then the antibiotic is:	Further action / remark:
Sensitive	<= (any number)	effective at the lowest concentration tested	should be effective choice
Sensitive	(any number)	effective, but not at the lowest concentration tested	refer to MIC range to determine where in the range the strain falls and compare this with other choices
Intermediate	(any number)	may be effective in high dosages or if it concentrates at the site of infection	refer to reference ranges for achievable antibiotic levels in affected tissues
Resistant	(any number)	will be unlikely to reach effective serum levels	choose an antibiotic that tests as sensitive
Resistant	>= (any number)	unlikely to be effective	choose an antibiotic that tests as sensitive

***In vitro* efficacy of pradofloxacin and erythromycin for a *Staphylococcus pseudintermedius* strain:**

Pradofloxacin, *Staph. pseudintermedius*



Erythromycin, *Staph. pseudintermedius*



For example: Erythromycin is 4 dilutions away from its breakpoint while pradofloxacin is only 3 dilutions away from its breakpoint. Therefore, in this case, although the absolute MIC value measured for pradofloxacin (0.25 µg/ml) is lower than the one measured for erythromycin (0.5 µg/ml), the strain is more susceptible to erythromycin.

Our consultants are available to help you interpret test results.

The introduction of MIC values enables a quantitative result, while the qualitative assessment is expressed as Sensitive, Intermediate or Resistant. This last classification, into effectiveness classes, is of direct clinical relevance to the veterinary practitioner.

The MIC value is primarily a parameter that is required for the precise evaluation of the laboratory test. It is required to establish the *in vitro* effectiveness of the antibiotic with respect to the isolate. As has already been mentioned in the example earlier, the MIC values of different agents for clinical use cannot be compared with each other directly. An antibiotic with an MIC value of 0.25 µg/ml can prove less effective than one with an MIC value of 0.5 µg/ml. This may seem illogical, since in the case of the latter agent, double the concentration seems to be required to inhibit microbial growth *in vitro*. However, when actually used on the patient, pharmacokinetic and clinical parameters also come into play. These factors were taken into account in the establishment of the clinical breakpoint by the CLSI and lead to the clinical classification of each isolate as sensitive, intermediate or resistant to an agent. Thus, an isolate may be graded sensitive to an agent at an equal or even higher MIC, while the same isolate has to be judged resistant to another agent with the same or even a lower MIC. This is one more demonstration that the MIC value or its level is insufficient on its own to permit any clinical pronouncement and, furthermore, has to be assessed individually, depending on agent and pathogen. No conclusion regarding the necessary therapeutic dose can be drawn directly from the MIC value. Only when the pharmacological parameters, such as tissue distribution, attainable concentration in the target tissue, rate of elimination, toxicity thresholds, etc. are precisely known can the MIC value be used for clinical applications. For direct clinical use, the therapeutic interpretation (S, I, or R) is the basis for successful therapy.

Class-reference antibiotics

Some antibiotics are used to determine the susceptibility of other antibiotics in the same class. For example, the presence of methicillin-resistant staphylococci (MRS) is tested in the laboratory with oxacillin and / or cefoxitin and not methicillin. The name MRS is used because of convention over years of use in scientific articles and textbooks.

Antimicrobial agent	Susceptibility prediction for:
Cephalexin	All first-generation cephalosporins, except cefazolin
Clindamycin	Lincosamides (lincomycin, pirlimycin) (GP only)
Erythromycin	Azithromycin and clarithromycin (GP only), macrolides (14), (in part) tylosin (16), spiramycin
Oxacillin	Methicillin (MRS) and isoxazolylpenicillines (cloxa-, dicloxacilline)
Tetracycline	Doxycycline, oxytetracycline
Sulfamethoxazol/ trimethoprim	Other potentiated sulfonamides
Ampicillin	Aminopenicillines (e.g. amoxicillin); penicillin (GP)
Kanamycin	Neomycin, framycetin, paromomycin
Chloramphenicol	Phenicoles (florfenicol)

(GP = Gram-positive microorganisms)

Antibiotic selection

When selecting an antibiotic, keep in mind that other factors in addition to the MIC are important. The location of the infection is important because lipid-soluble drugs reach higher levels in the tissue than they do in serum. Drugs excreted by the kidney reach much higher bladder levels than serum levels. Also, some drugs are more effective against Gram-negative bacteria than Gram-positive bacteria and vice versa. Species considerations are also important because certain antibiotics are toxic in some species.

Therefore, the choice of antibiotic should be based not on the MIC alone but on several other criteria.

Factors for selecting the most appropriate agent include:

- The MIC numerical value
- The antibiotic's breakpoint (how far the MIC is from the breakpoint)
- The site of infection (achievable drug levels at site of infection)
- Age of patient, animal species, health of animal (known organ disorders, immune status)
- Mode of action (bacteriostatic / bactericide), spectrum (broad-spectrum versus narrow-spectrum)
- Pharmacokinetics (volume of distribution, bioavailability, route of elimination)
- Restrictions in application (antibiotic guidelines according to countries, or restrictions for use in farm animals, restrictions for critically important antibiotics)
- Safety / contraindications, possible side effects of the drug, toxicity margin
- Ease of use, frequency and route of administration

Consider the appropriate criteria in each clinical case when determining the optimum antibiotic.

For the choice of an antibiotic one has also to take into consideration that different countries have different antibiotic guidelines. Regional recommendations and legal restrictions should also be taken into consideration. Antibiotics considered critically important should only be used with great care and never as first-line drugs.

And finally: why is sometimes no susceptibility report available at all?

At IDEXX we follow international guidelines set by the Clinical Laboratory Standards Institute (CLSI) combined with our years of experience in performing susceptibility testing. As such the following is an outline of our antibiotic testing policy:

- Susceptibilities are not performed on normal flora and non-pathogenic microorganisms.
- Pathogens with no international interpretative standards will be reported with a recommended list of antibiotics (if available from scientific sources). Examples include *Corynebacterium* species and *Campylobacter* species.
- In case susceptibility testing is not possible (e.g. method no applicable, general or individual growth failure) a recommended list of antibiotics will be reported (if available from scientific sources).
- Topical agents will not be tested. There are NO international guidelines for laboratory-based susceptibility testing of these agents. Any testing of such may lead to serious misleading results. Examples include agents included in eye and ear preparations.
- We do not test inappropriate organism/antibiotic combinations, such as Clindamycin against Gram-negative organisms or penicillin against *E. coli* (both examples have intrinsic resistance mechanisms against these antibiotics).

Customer support services

IDEXX supports your practice with our customer support, technical support, and medical consulting services teams, including our diagnostic support veterinarians and veterinary specialists.

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