A Proposed Evidence-Based Local Guideline for Definition of Multidrug-Resistant (MDR), Extensively Drug-Resistant (XDR) and Pan Drug-Resistant (PDR) Bacteria by the Microbiology Laboratory

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ABSTRACT: Multi-drug resistant organisms (MDROs) are defined as microorganisms, predominantly bacteria, that are resistant to one or more classes of antimicrobial agents. These pathogens are frequently resistant to most available antimicrobial agents and deserve special attention in healthcare facilities. Generally, MDRO infections have clinical manifestations similar to infections caused by susceptible pathogens. Despite of two different classifications by WHO and CDC, still there are debates about update definition of MDROs in medical literature. Here we provide an updated local guideline for definitions of various MDROs by microbiology laboratory.

KEY WORDS: Multi-drug resistant organisms (MDROs), Antimicrobial agent, Healthcare facilities, microbiology laboratory

Worldwide emerging data suggest that multidrug resistant organisms (MDROs) have considerably increased in the recent decades. The worldwide spread of organisms that are resistant to multiple antibiotics, generally referred to as multidrug-resistant organisms (MDRO), has become a public health concern. (1,2) Although the names of certain multi-drug resistant organisms (MDROs) describe resistance to only one agent as in methicillin-resistant staphylococcus aureus (MRSA) and vancomycin resistant enterococcus (VRE), these pathogens are frequently resistant to most available antimicrobial agents. These highly resistant organisms deserve special attention in healthcare facilities. In addition to MRSA and VRE, certain gram-negative bacteria (GNB), including extended spectrum beta-lactamases-producing Enterobacteriaceae (ESBL-PE), Carbapenem-resistant Pseudomonas aeruginosa & Acinetobacter baumannii (CRPA & CRAB), other carbapenem-resistant Enterobacteriaceae (CRE), also Stenotrophomonas maltophilia and Burkholderia cepacia that are intrinsically resistant to the broadest-spectrum antimicrobial agents are now emerging worldwide. In most instances, MDRO infections have clinical manifestations that are similar to infections caused by susceptible pathogens. However, options for treating patients with these infections are usually limited. Increased lengths of stay, costs, and mortality also have been associated with MDROs. Among MDROs, MRSA is differently from other MDROs. When patients with MRSA have been compared to patients with methicillin-susceptible S. aureus (MSSA), MRSA-colonized patients more frequently develop symptomatic infections. Furthermore, higher case fatality rates have been observed for certain MRSA infections, including bacteremia. Staphylococcus aureus, vancomycin-intermediate (VISA) and resistant (VRSA) are also difficult to treat emerging variants.

The main sources of information on rates of infection caused by MDROs come from regional, national or international surveillance systems, published epidemiological studies and outbreak reports. On 2017, WHO published its first list of antibiotic-resistant "priority pathogens" (12 families of bacteria that pose the greatest threat to human health). (3) The criteria for selecting pathogens on the list were:

1. How deadly the infections they cause are; whether their treatment requires long hospital stays
2. How frequently they are resistant to existing antibiotics when people in communities catch them
3. How easily they spread between animals, from animals to humans, and from person to person
The list highlights in particular the threat of gram-negative bacteria that are resistant to multiple antibiotics. The WHO list is divided into three categories according to the urgency of need for new antibiotics: critical, high and medium priority. (3) (Table 1)

The most critical group of all includes multidrug resistant bacteria that pose a particular threat in hospitals, nursing homes, and among patients whose care requires devices such as ventilators and blood catheters. They include Acinetobacter, Pseudomonas and various Enterobacteriaceae (including Klebsiella, E. coli, Serratia, and Proteus). They can cause severe and often deadly infections such as bloodstream infections and pneumonia. These bacteria have become resistant to a large number of antibiotics, including carbapenems and third generation cephalosporins – the best available antibiotics for treating multi-drug resistant bacteria. The second and third tiers in the list – the high and medium priority categories – contain other increasingly drug-resistant bacteria that cause more common diseases such as gonorrhea and food poisoning caused by salmonella.

Tuberculosis was not included in the list. Other bacteria that were not included, such as streptococcus A and B and chlamydia, have low levels of resistance to existing treatments and do not currently pose a significant public health threat.

On the other hand, CDC is also concerned about rising resistant infections in the community, which can put more people at risk, make spread more difficult to identify and contain, and threaten the progress made to protect patients in healthcare. The emergence and spread of new forms of resistance remains a concern. On 2019, CDC released the lists of 18 antibiotic-resistant bacteria and fungi into three categories based on level of concern to human health—urgent, serious and concerning. (4) The report also includes a Watch List with three threats that have not spread resistance widely in the U.S. but could become common without a continued aggressive approach. (4) (Table 2)

A resistant interpretation of an isolate can be determined using disk diffusion, broth microdilution or agar dilution following Clinical and Laboratory Standards Institute (CLSI) guidelines for susceptibility testing and interpretation of Enterobacteriaceae, P. aeruginosa and Acinetobacter spp. (5) Current CLSI M100 breakpoints should be used to determine antimicrobial susceptibility of isolates (5). MIC or zone diameter value breakpoints and interpretive categories are established per CLSI document M235 for categories of susceptible, intermediate, and resistant (and susceptible-dose dependent and non-susceptible, when appropriate).

- **Susceptible (S)** – a category defined by a breakpoint that implies that isolates with an MIC at or below or a zone diameter at or above the susceptible breakpoint are inhibited by the usually achievable concentrations of antimicrobial agent when the dosage recommended to treat the site of infection is used, resulting in likely clinical efficacy.

- **Susceptible-dose dependent (SDD)** – a category defined by a breakpoint that implies that susceptibility of an isolate depends on the dosage regimen that is used in the patient. To achieve levels that are likely to be clinically effective against isolates for which the susceptibility testing results (either MICs or zone diameters) are in the SDD category, it is necessary to use a dosage regimen (ie, higher doses, more frequent doses, or both) that results in higher drug exposure than that achieved with the dose that was used to establish the susceptible breakpoint. Consideration should be given to the maximum, literature-supported dosage regimen, because higher exposure gives the highest probability of adequate coverage of an SDD isolate. Appendix E lists the doses used when establishing SDD categories. The drug label should be consulted for recommended doses and adjustment for organ function. (NOTE: The SDD category may be assigned when doses well above those used to calculate the susceptible breakpoint are supported by the literature, widely used clinically, and/or approved and for which sufficient data to justify the designation exist and have been reviewed.)

- **Intermediate (I)** – a category defined by a breakpoint that includes isolates with MICs or zone diameters within the intermediate range that approach usually attainable blood and tissue levels and/or for which response rates may be lower than for susceptible isolates. (NOTE: The intermediate category implies clinical efficacy in anatomical sites where the drugs are physiologically concentrated.)

- **Resistant (R)** – a category defined by a breakpoint that implies that isolates with an MIC at or above or a zone diameter at or below the resistant breakpoint are not inhibited by the usually achievable concentrations of the agent with normal dosage schedules.
and/or that demonstrate MICs or zone diameters that fall in the range in which specific microbial resistance mechanisms are likely, and clinical efficacy of the agent against the isolate has not been reliably shown in treatment studies.

- **Non-susceptible (NS)** – a category used for isolates for which only a susceptible breakpoint is designated because of the absence or rare occurrence of resistant strains. Isolates for which the antimicrobial agent MICs are above or the zone diameters are below the value indicated for the susceptible breakpoint should be reported as non-susceptible. *(NOTE 1: An isolate that is interpreted as Non-susceptible does not necessarily mean that the isolate has a resistance mechanism. It is possible that isolates with MICs above the susceptible breakpoint that lacks resistance mechanisms may be encountered within the wild-type distribution after the time the susceptible-only breakpoint was set; NOTE 2: The term “non-susceptible” should not be used when the text is describing an organism/drug category with intermediate and resistant interpretive categories.)*

Some laboratories may routinely use other breakpoint interpretations such as European Committee on Antimicrobial Susceptibility Testing (EUCAST) that differ from CLSI recommendations. Laboratories using non-CLSI breakpoints, should disclose this information in their reports to provincial public health laboratories. Certain species of Enterobacteriaceae should not be tested for particular antimicrobial agents because of intrinsic resistance. (for example, Proteus spp. are intrinsically resistant to tigecycline and colistin while Serratia spp. are naturally resistant to Colistin)

On the other hand, recently EUCAST has changed the definitions of susceptibility testing categories S, I and R: (6)

- **S – Susceptible:** A microorganism is categorized as "Susceptible, standard dosing regimen", when there is a high likelihood of therapeutic success using a standard dosing regimen of the agent.

- **I - Susceptible, increased exposure:** A microorganism is categorized as "Susceptible, Increased exposure when there is a high likelihood of therapeutic success because exposure to the agent is increased by adjusting the dosing regimen or by its concentration at the site of infection.

- **R - Resistant:** A microorganism is categorized as "Resistant" when there is a high likelihood of therapeutic failure even when there is increased exposure.

Despite of these two different classifications by WHO and CDC, still there are debates about update definition of MDROs in medical literature. Wrong or old definitions for MDROs, especially regarding GNBs, will be misleading and can cause false epidemiologic statistics and clinical decision making. CDC typically uses MDRO to refer to an isolate that is resistant to at least one antibiotic in three or more drug classes. (4)

According to Canadian recommendations for laboratory interpretation of multiple or extensive drug resistance in clinical isolates of Enterobacteriaceae, Acinetobacter species and Pseudomonas aeruginosa there is only definition for MDR Enterobacteriaceae and XDR Acinetobacter spp. or P. aeruginosa definition. (7) Regarding Enterobacteriaceae, an isolate should be considered a MDRO if it is resistant to THREE OR FOUR of the SIX antimicrobial groups including Tobramycin OR gentamicin (except for Serratia spp.), Piperacillin-tazobactam, Imipenem OR meropenem (except for Proteus spp.), Cefotaxime OR ceftriaxone OR ceftazidime, Ciprofloxacin and Trimethoprim-sulfamethoxazole. However, about Acinetobacter spp. or P. aeruginosa, there are no final recommendations for MDRO definitions for Acinetobacter spp. or P. aeruginosa. The previous interim recommendations for Acinetobacter spp. or P. aeruginosa MDRO status should be disregarded at this time (8). An isolate should be considered an XDRO if it is resistant to All of the five antimicrobial groups including Ciprofloxacin, Piperacillin-tazobactam, Ceftazidime, Imipenem or meropenem and Tobramycin. The most important pitfall of this classification is lack of definition for MDR Acinetobacter spp. or P. aeruginosa. Also, they have no idea about XDR and PDR pathogens.

We preferred to define MDR, XDR and PDR according to the “international expert proposal for interim standard definitions for acquired resistance” (9). MDR refers to strains that exhibit resistance to more than three or more antimicrobial drug classes (9,10). XDR refers to strains resistant to all but two drug classes (9,11). Pan-drug resistance refers to resistance exhibited by the strains to all drug classes (9,12) Thus, we reviewed all new studies regarding definition and reporting of MDROs (13-24) and have created an updated, local guideline for definition for MDR, XDR and PDR pathogens which is scientific and practical for all microbiology laboratories at national level. (Table 3)
In conclusion, although till date there was no comprehensive consensus regarding detailed definitions of MDR, XDR and PDR pathogens have been proposed, however, our new practical definition according to latest evidences can help microbiology laboratories for report and infectious disease physicians to deal better with MDROs in practice.

Table 1: WHO priority pathogens list

| Priority 1: CRITICAL | Priority 2: HIGH | Priority 3: MEDIUM |
|---------------------|-----------------|-------------------|
| *Carbapenem-resistant Acinetobacter baumannii | *Vancocmycin resistant enterococcus | *Streptococcus pneumoniae, penicillin-non-susceptible |
| *Carbapenem-resistant *Pseudomonas aeruginosa | *Methicillin-resistant, vancocmycin intermediate & vancocmycin resistant Staphylococcus aureus | *Hemophilus influenzae, ampicillin-resistant |
| *Carbapenem-resistant Enterobacteriaceae | *Campylobacter spp., fluoroquinolone-resistant | *Shigella spp., fluoroquinolone-resistant |
| *ESBL-Producing Enterobacteriaceae | *Salmonellae, fluoroquinolone-resistant | |
| *Neisseria gonorrhoeae, cephalosporin-resistant, fluoroquinolone-resistant | | |

Table 2: CDC 2019 ANTIBIOTIC Resistance (AR) Threats Report

| Urgent Threats | Serious Threats | Concerning Threats | Watch List |
|----------------|-----------------|-------------------|------------|
| *Carbapenem-resistant Acinetobacter baumannii | *Drug-resistant Campylobacter | *Erythromycin-Resistant Group A Streptococcus | *Azole-resistant Aspergillus fumigatus |
| *Candida auris | *Drug-resistant Candida | *Clindamycin-resistant Group B Streptococcus | *Drug-resistant Mycoplasma genitalium |
| *C. difficile | *ESBL-Producing Enterobacteriaceae | | *Drug-resistant Bordetella pertussis |
| *Carbapenem-resistant Enterobacteriaceae | *Vancocmycin resistant enterococcus | | |
| *Drug-resistant Neisseria gonorrhoeae | *MDR P. aeruginosa | | |
| | *Drug-resistant S. Typhi & nontyphoidal Salmonella | | |
| | *Drug-resistant Shigella | | |
| | *Methicillin-resistant Staphylococcus aureus | | |
| | *Drug-resistant S. pneumoniae | | |
| | *Drug-resistant Tuberculosis | | |

Table 3: Proposed new definition of MDR, XDR, PDR

| Pathogen(s) | Must be tested | MDR | XDR | PDR |
|-------------|----------------|-----|-----|-----|
| *Staphylococcus aureus* | *Gentamicin* | non-susceptible to ≥ 1 agent in ≥ 3 antimicrobial categories. | non-susceptible to ≥ 1 agent in all but ≤ 2 categories. (Only susceptible to 1-2 remaining agent) | non-susceptible to all antimicrobial agents listed. |
| | *Oxacillin* | | | |
| | *Ciprofloxacin* | | | |
| | *TMP/SMX* | | | |
| | *Vancomycin or Teicoplanin* | | | |
| | *Tigecycline* | | | |
| | *Erythromycin* | | | |
| | *Clindamycin* | | | |
| | *Linezolid* | | | |
| | *Doxycycline or Minocycline* | | | |
| | *Rifampicin* | | | |
| **Enterococcus faealis** | *Gentamicin & Streptomycin (high level)*  
*Imipenem or Meropenem*  
*Vancomycin or Teicoplanin*  
*Tigecycline*  
*Ampicillin*  
*Linezolid*  
*Doxycycline*  
*Ciprofloxacin* | Same as above | Same as above | Same as above |
|------------------------|--------------------------------------------------|----------------|----------------|----------------|
| **Enterococcus faecium** | *Gentamicin & Streptomycin*  
*Vancomycin or Teicoplanin*  
*Tigecycline (when no other option)*  
*Ampicillin*  
*Linezolid*  
*Doxycycline*  
*Ciprofloxacin* | Same as above | Same as above | Same as above |
| **E. coli** | *Amoxicillin-clavulanate*  
*Amikacin or Gentamycin*  
*Piperacillin/tazobactam*  
*Imipenem or Meropenem*  
*Cefazolin or Cefuroxime*  
*Cefotaxime or Ceftiraxone or Ceftazidime*  
*Ciprofloxacin*  
*Trimethoprim-sulfamethoxazole*  
*Doxycycline*  
*Tigecycline (when no other option)*  
*Colistin (when no other option)* | Same as above | Same as above | Same as above |
| **Klebsiella spp.** | *Amoxicillin-clavulanate*  
*Amikacin or Gentamycin*  
*piperacillin/tazobactam*  
*Imipenem or Meropenem*  
*Cefazolin or Cefuroxime*  
*Cefotaxime or Ceftiraxone or Ceftazidime*  
*Ciprofloxacin*  
*Trimethoprim-sulfamethoxazole*  
*Doxycycline*  
*Tigecycline (when no other option)*  
*Colistin (when no other option)* | Same as above | Same as above | Same as above |
| **Providencia spp.** | *Amikacin*  
*piperacillin-tazobactam*  
*Imipenem or Meropenem*  
*Cefuroxime*  
*Cefotaxime or Ceftiraxone or Ceftazidime*  
*Ciprofloxacin*  
*Trimethoprim-sulfamethoxazole*  
[Doxycycline*  
*Tigecycline (when no other option)*  
*Colistin (when no other option)* | Same as above | Same as above | Same as above |
| **Citrobacter spp. (Freundi, Koseri,…)** | *Amikacin or Gentamycin*  
*piperacillin-tazobactam*  
*Imipenem or Meropenem* | Same as above | Same as above | Same as above |
| **Enterobacter aerogenes** & **E. cloacae** | *Cefuroxime*<br>*Cefotaxime or Ceftriaxone or Ceftazidime*<br>*Ciprofloxacin*<br>*Trimethoprim-sulfamethoxazole*<br>*Doxycycline*<br>*Tigecycline (when no other option)*<br>*Colistin (when no other option)* | Same as above | Same as above | Same as above |
| **Morganella morganii** | *Cefuroxime*<br>*Cefotaxime or Ceftriaxone or Ceftazidime*<br>*Ciprofloxacin*<br>*Trimethoprim-sulfamethoxazole*<br>*Doxycycline*<br>*Tigecycline (when no other option)*<br>*Colistin (when no other option)* | Same as above | Same as above | Same as above |
| **Serratia marcescens** | *Cefuroxime*<br>*Cefotaxime or Ceftriaxone or Ceftazidime*<br>*Ciprofloxacin*<br>*Trimethoprim-sulfamethoxazole*<br>*Doxycycline*<br>*Tigecycline (when no other option)* | Same as above | Same as above | Same as above |
| **Non Mirabilis Proteus** (*Vulgaris, Penneri,..*) | *Cefuroxime*<br>*Cefotaxime or Ceftriaxone or Ceftazidime*<br>*Ciprofloxacin*<br>*Trimethoprim-sulfamethoxazole*<br>*Doxycycline*<br>*Tigecycline (when no other option)* | Same as above | Same as above | Same as above |
| **Proteus Mirabilis** | *Cefuroxime*<br>*Cefotaxime or Ceftriaxone or Ceftazidime*<br>*Ciprofloxacin*<br>*Trimethoprim-sulfamethoxazole*<br>*Doxycycline*<br>*Tigecycline (when no other option)* | Same as above | Same as above | Same as above |
### Pseudomonas aeruginosa
*Amikacin or Gentamycin
*piperacillin-tazobactam
*Imipenem or Meropenem
*Ceftazidime or Cefepime
*Ciprofloxacin or Levofoxacin
*Colistin (when no other option)

Same as above
Same as above
Same as above

### Acinetobacter spp. (including A. baumannii)
*Amikacin or Gentamycin
*Imipenem or Meropenem
*Cefotaxime or Ceftriaxone or Ceftazidime
*Ciprofloxacin or Levofoxacin
*Trimethoprim-sulfamethoxazole
*Doxycycline or Minocycline
*Tigecycline (when no other option)
*Colistin (when no other option)

Same as above
Same as above
Same as above

### Stenotrophomonas maltophilia
*Ceftazidime
*Levofoxacin
*Trimethoprim-sulfamethoxazole
*Tigecycline (when no other option)
*Colistin (when no other option)
*Minocycline or Doxycycline

Same as above
Same as above
Same as above

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