

# Interim Proposal for Surveillance of Multidrug-Resistant, Extensively Drug-Resistant and Pandrug-Resistant Bacteria for the Automated AST User Centres

Sastry Apurba Sankar<sup>1\*</sup>, Priyadarshi Ketan<sup>1</sup>, Rajashekar Deepashree<sup>2</sup>, and Rai Sumit<sup>3</sup>

<sup>1</sup>Department of Microbiology, JIPMER, Puducherry, India

<sup>2</sup>Department of Microbiology, JSS Medical college, Mysore, India

<sup>3</sup>Department of Clinical Microbiology and Infectious Diseases, Super Speciality Paediatric Hospital and PG Teaching Institute, Noida, India

## Abstract

In the era of increased Antimicrobial Resistance (AMR), it is important that Healthcare Facilities (HCFs) should conduct surveillance to know the true estimate of drug resistant organisms such as Multidrug-Resistant (MDR), Extensively Drug-Resistant (XDR) and pandrug-resistant (PDR) bacteria prevalent in their facility. The limitations of disk diffusion method used for comparison of AMR data are largely overcome by use of MIC based automated AST method. These systems use panels comprising of fixed set of large number of antimicrobials and provide standard testing protocol which remains uniform across the user HCFs. The use of automated AST systems has been increasing in the recent past and is expected to expand further in future. Among the automated AST systems available, VITEK-2 is the most extensively used platform both globally (~63%) and in India (~85%). This study provides the guideline to develop revised templates comprising of antimicrobial classes and agents based on automated AST panels for the purpose of MDR/XDR/PDR categorization. By using these templates, AMR data of various automated AST user centres can be collated to achieve meaningful comparison of AMR data. Furthermore, there is no need for any additional manpower or budget, as the analysis of drug resistant bacteria is performed based on routine AST data, without any additional testing. Therefore, large number of HCFs can contribute their AMR data, which can be collated to give true picture of the current burden of MDR/XDR/PDR at national and global level. This information is essential for developing empirical antimicrobial therapy for diverse epidemiological settings.

**Keywords:** MDR • XDR • PDR • AMR surveillance • Antimicrobial resistance • Automated AST • VITEK

## Introduction

Antimicrobial Resistance (AMR) represents a leading menace to the global public health, contributing to a significant morbidity and mortality, with substantial economic impact. It is of particular concern in resource-constrained countries like India, where the burden of infectious diseases is very high and the consumption of antimicrobials is massive and un-regulated. Consistent and reliable estimate of the true AMR burden in a healthcare facility (HCF) is the cornerstone for providing information on local resistance patterns, to monitor the AMR trend across different time frames, and for inter-institutional comparison. It is also useful for evaluating the effectiveness of AMR containment interventions such as Antimicrobial Stewardship (AMSP) initiatives, infection prevention and control measures, and formulating the institutional empirical antimicrobial policy.

The three important terminologies that are commonly used to characterize the different AMR patterns found in healthcare-associated pathogens include Multidrug-Resistant (MDR), Extensively Drug-Resistant (XDR) and Pandrug-Resistant (PDR) bacteria. Although there are many diverse classifications described in the literature, the interim guideline proposed by the joint ECDC/CDC expert group [European Centre for Disease Prevention and Control (ECDC) and Centre for Disease Prevention and Control (CDC)] is internationally the most accepted definition to characterize MDR, XDR and PDR bacteria. Though the definitions used here are clear and precise, the list of antimicrobial classes and agents recommended for testing to meet to the criteria of MDR, XDR and PDR definitions is exhaustive and may not be available in all the facilities. This serves as an important barrier for accurately classifying the drug-resistant bacteria in the Healthcare Facilities (HCF), a major hindrance for inter-institutional comparison of AMR data [1].

\*Address to correspondence: Sastry Apurba Sankar, Department of Microbiology, JIPMER, Puducherry, India, Tel: 919444327314; E-mail: apurbasastryresearch@gmail.com

**Copyright:** © 2021 Sankar SA, et al. This is an open-access article distributed under the terms of the creative commons attribution license which permits unrestricted use, distribution and reproduction in any medium, provided the original author and source are credited.

**Received:** 06 October, 2021; **Accepted:** 20 October, 2021; **Published:** 27 October, 2021

## Rationale

In order to accomplish reliable epidemiological surveillance data on the frequency distribution of the drug-resistant bacteria, which can be used to achieve a true comparison of inter-institutional AMR burden, HCFs must employ the identical method of performing Antimicrobial Susceptibility Testing (AST) with a fixed set of antimicrobial agents and classes and harmonized definitions to characterize the drug-resistant bacteria. In Indian facilities, the AST is carried out mainly by two methods. While the conventional Disk Diffusion (DD) test still accounts for the most common AST method employed in the majority of Indian laboratories use of automated AST methods has exponentially increased in many clinical laboratories in the recent past. The comparison of data of MDR, XDR, and PDR bacteria between the centres that use DD method has always been found to be difficult, which may be attributed to several factors (i) diverse list of antimicrobials used in DD testing by different centres, depending up on their local practices, (ii) differences in the protocols used to perform DD test (e.g. culture medium used, use of disks from different manufacturers, and varied incubatory conditions (such as temperature, duration of incubation), (iii) inter-observer variations in measuring zone diameter, (iv) manual validation of result leading to variation in interpretation, and (v) and disparity in quality control etc.s

The limitations of DD methods are largely overcome by the use of automated MIC based AST method. There are primarily three major automated AST systems available worldwide-VITEK-2, Phoenix and MicroScan. These systems work on the principle of broth microdilution and obtain the AST result based on Minimum Inhibitory Concentration (MIC), which is more reliable and accurate than the zone diameters of DD method. These systems employ antibiotic panels, comprising of standard set of antimicrobial agents, therefore brings the uniformity of the list of antimicrobial used among the user centres. These systems generally have a standard protocol for performing the test and the adequate training is provided to the user centres by the manufacturer, which overcomes the problem of non-uniformity of protocol across the HCFs, as observed in DD method. The manufacturer usually provides a competency evaluation at the end of the training sessions. Among the automated AST systems, VITEK-2 is the most extensively used platform both globally (~63%) and also in Indian settings (~85%). There are more than 1200 users of this the automated AST system in India. If a revised template for MDR/XDR/PDR categorization is available comprising of antimicrobial classes and agents based on the automated AST panels, then the AMR data of various automated AST user centres can be collated to achieve a meaningful comparison; thus the burden of MDR, XDR and PDR of HCFs across the countries can be determined. Therefore, this study was undertaken to develop an interim proposal for surveillance of multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria for the automated AST user centres [2].

## Methodology

### Criteria used to include antimicrobial agents

For the purpose of classifying drug resistant bacteria, the authors have formulated the templates comprising of lists of antimicrobial agents and classes tested for each organism group. Only those

antimicrobial agents, that are included in the respective automated AST panel are considered while constructing the templates. The development of these templates based on four commonly used AST panels in VITEK-2 (i) AST N280/N405 panel (for Enterobacterales),

(ii) AST N281/N406 panel (for non-fermenters), (iii) AST P628 panel (for Staphylococcus/Enterococcus group), and (iv) AST ST03 panel (for Streptococcus group). The inclusion or exclusion of an agent to the antimicrobial template for classifying drug resistant bacteria have been based on certain set rules. The antimicrobial agent can be included to an organism's antimicrobial template only when (i) it is listed in the respective automated AST panels and tested against the corresponding organism, (ii) clinical breakpoints are available, and (iii) it is therapeutically used in the treatment of that particular organism (clinically indicated).

A standard approach has been proposed for the laboratories to use internationally accepted clinical breakpoints while interpreting the AST result. CLSI being the most popular and widely used guideline for AST, the clinical breakpoints of CLSI should be applied whenever possible. For those drug/bug combinations for which CLSI breakpoints are not available, the laboratories can use the clinical breakpoints of EUCAST. In case the EUCAST breakpoints are also not available, then any other clinical breakpoints (such as FDA) can be used. In the antimicrobial templates given in Table 1-4, the type of clinical breakpoints to be used for each drug/bug combination is clearly mentioned, and the centres who intend to compare their AMR data with others should implement the same in their laboratories. As per the general principle of antibiogram, the susceptibility result of only the first isolate of an organism encountered in a given patient should be included for analysis [3].

Similarly, an antimicrobial agent should be considered to be excluded from the template of an organism if: (i) the organism is found to be intrinsically resistant (IR) to that antimicrobial agent; or (ii) clinical breakpoints are not available (BPN); or (iii) There has been applied any restriction rule to release the result of the MIC as well as the interpretation (e.g. *Stenotrophomonas/ceftazidime*) by manufacturer or (iv) clinical breakpoints are available, but therapeutically are not used (e.g. meropenem for *Salmonella*), or (v) only epidemiological cut off breakpoints are available, but not clinical breakpoint (*Pseudomonas/fosfomycin*), or (vi) non-susceptible results if obtained, it has to be re-identified, retested and if confirmed, then submitted to a reference laboratory.

The site-specific antimicrobials although are included in the template, they will be considered for analysis only for the site specific isolates and will be excluded from the analysis for the isolates recovered from other specimens. For example, antimicrobials for which urine-only breakpoints are available and achieve therapeutic concentrations only in urine (e.g. nitrofurantoin) will be considered for analysis for the urinary isolates only, and will be excluded for non-urinary isolates. Similarly, daptomycin as is not reported for respiratory isolates and therefore will only be included for analysis of non- respiratory isolates.

### Quality of the testing method

The reliability of automated AST report essentially depends upon the quality of the testing method. The laboratories must strictly adhere to the protocol provided by the manufacturer. Common error prone steps where extreme care needs to be taken include picking of

single type of isolated colonies, adjusting the inoculum to 0.5 McFarland by densitometer (DensiChek) and loading the panel within 15-20 min of preparation. The antimicrobials which are flagged as 'terminated' will be considered as 'not tested' during analysis. The Quality Control (QC) of automated AST should routinely be performed according to CLSI M07, 11<sup>th</sup> edition; the frequency can either be weekly or on daily basis, depending up on the laboratory usage. Additional QC testing should be performed when a new shipment/lot of AST panel is procured or change in antimicrobial dilution.

### Data validation

The AST data from automated instrument must be validated by the clinical microbiologist before inclusion for analysis. The laboratories may not always include certain antimicrobials in patient's report because of local antibiotic practice or drug unavailability; for e.g., teicoplanin for *S. aureus* may not be included in clinical reports of most Indian laboratories because of unavailability. However as a routine, the laboratories should develop a practice of verifying the AST data of all the antimicrobials for which automated AST results are available and subsequently validating the result, regardless of inclusion of antimicrobials in patient's final report. The instrument provides an automated tool to validate AST results called 'Advanced Expert System (AES)'. It ensures the quality of AST test results and decreases the chance for human error through rapid, automatic and systematic validation of every susceptibility test result. Each MIC result is checked against a database of more than 3,500 phenotypes and 30,000 MIC distributions to determine consistency with previously defined wild or resistant phenotypes. After AES validation, the susceptibility results are released by the automated AST instrument with an additional comment as 'consistent result', or 'inconsistent result', or 'consistent result with modification'.

AST data with 'consistent result' indicates isolates fit an expected pattern for a defined phenotype; and therefore can be included for analysis. The AST data that are flagged by AES as 'consistent with modification' indicates that the MIC values do not fit the expected pattern for a known phenotype. Such results should be verified by the clinical microbiologist and added for analysis only after their validation. The AST data with inconsistent result should be excluded from analysis, as it indicates that the MIC pattern for the tested isolate could not be matched to a known phenotype in the software database. The software provided with the automated AST instrument has provision to invoke selective reporting rules, which the laboratories can apply to suppress certain susceptibility results from the patient report. In such case, the laboratories must disable the suppression rules, before downloading the AST data for the purpose of analysis of drug resistant bacteria. It is recommended that all final verified test results, including those that might be suppressed on patient report should be added for analysis [4].

For suspicious AST report for some drug/bug combinations, it is recommended to re-confirm the data by an additional testing method such as disk diffusion or Epsilon meter test. Some examples of such suspicious AST report for which a reconfirmation is definitely advisable include: vancomycin resistance for *S. aureus*, oxacillin and cefotaxim for *S. aureus* (when contradictory results are produced), colistin resistance, linezolid resistance in *Enterococcus* and *Staphylococcus* and isolated carbapenem

### Criteria used for defining antimicrobial classes

There has been no common consensus for determining the classes of antimicrobial agents that should be used for defining MDR, XDR and PDR. Often, a combination of the different approaches is to define the antimicrobial classes (i) chemical structures for antimicrobial classes (e.g. cephalosporins), (ii) antimicrobial subclasses, (e.g. first-generation cephalosporins) or organism-specific antimicrobial agents (e.g. antipseudomonal  $\beta$ -lactams). Even the ECDC/CDC expert group also used combination of the above three approaches to define antimicrobial classes [5]. This combinational approach creates ambiguity and non-uniformity, which makes it challenging to compare the results between different studies. The authors in this review, therefore recommend to use only one approach (i.e. based on chemical structure) for defining the antimicrobial classes, adapted from CLSI M100 31<sup>st</sup> edition. Use of single-approach based classification of antimicrobial class will bring homogeneity, and will facilitate a meaningful comparison of the MDR/XDR/PDR data between the centres.

Only three noteworthy exceptions can be made to the CLSI's list of antimicrobial classes-(i) anti-staphylococcal beta-lactams (e.g. oxacillin or ceftazidime) need to be used as a separate class as it is surrogate marker, exclusively used for defining methicillin resistant *Staphylococcus aureus* (MRSA); (ii) as tigecycline differs from other tetracyclines both in chemical structure and antimicrobial spectrum, it can be grouped into a separate class glycylicyclines; (iii) daptomycin and colistin differ from each other both in chemical structure and antimicrobial spectrum and therefore can be grouped separately into two distinct classes lipopeptides and polymyxins respectively. After applying the above mentioned criteria, the authors proposed for a total of 21 classes of antimicrobial agents. However, the exact number of classes for each organism will depend up on the antimicrobial agents included in their respective templates after applying the exclusion criteria.

### Organism specific templates

For the purpose of classifying drug resistant bacteria into MDR, XDR and PDR, the authors have designed organism specific templates comprising of antimicrobial agents (and their classes). There are four such templates (Tables 1-4) developed, each represents an organism group for which a specific automated AST panel is used for performing AST-(i) Enterobacterales (AST panel N280), (ii) Non-fermenter group (AST panel N281), (iii) *Staphylococcus/Enterococcus* group (AST panel P628) and (iv) *Streptococcus* group (AST panel ST03).

### Template for enterobacterales

The AST panel N280 is recommended for testing for Enterobacterales group. It comprises of a total of 18 antimicrobial agents from 10 different classes. The updated versions of this panel, AST panel N405 is available for use from June 2021 in India. The changes include addition of fosfomycin, instead of ampicillin; therefore the template needs to be adjusted accordingly. However, the exact number of antimicrobial agents and classes to be included in the template will differ according to the organism and clinical specimen. This panel contains a urinary antimicrobial, Nitrofurantoin; which should be included for analysis of only urinary isolates. Ampicillin being intrinsic resistant, can be excluded from analysis of

*Klebsiella pneumoniae*; whereas for *Enterobacter* species, both ampicillin and amoxicillin-clavulanate are excluded (intrinsic resistant). For *Salmonella*, the antimicrobials that are used therapeutically are included for analysis-ampicillin, ceftriaxone, trimethoprim-sulfamethoxazole and ciprofloxacin. In addition, the centres need to perform disk diffusion for chloramphenicol and azithromycin against *Salmonella*, which are not available in AST panel 280. The interpretation of AST result can be carried out by applying CLSI clinical breakpoints for most drug/bug combinations, except for fosfomycin and tigecycline, for which EUCAST and FDA clinical breakpoints may be applied.

**Template for staphylococcus/enterococcus group**

Template for *Staphylococcus/Enterococcus* group is illustrated in Table-3, which is developed based on AST panel P628. It comprises of a total of 17 antimicrobial agents from 14 different classes; however, their inclusion in the template will differ among the organisms based on the inclusion and exclusion criteria. The AST results can be interpreted by applying CLSI clinical breakpoints for most drug/bug combinations, except for fosfomycin and tigecyclines, for which EUCAST clinical breakpoints may be applied. This panel contains certain antimicrobials which should be included for analysis of only urinary isolates-nitrofurantoin (for *Staphylococcus* and *Enterococcus*); tetracycline, ciprofloxacin and levofloxacin (for *Enterococcus*). For *Enterococcus*, additional disk diffusion test can be performed for certain important antimicrobials which are not available in P628 panel such as ampicillin. Daptomycin can be included for analysis of non-respiratory isolates of *Staphylococcus*, and *E. faecalis*. High level gentamicin should be included in the template of *Enterococcus* only.

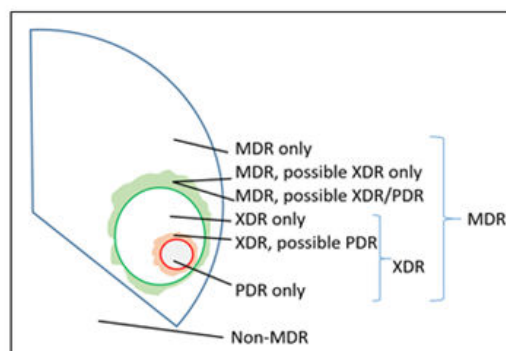
**Template for streptococcus group**

Table-4 depicts the template for *Streptococcus* group using AST panel ST03; which comprise of three major organism group-β haemolytic streptococci, *Streptococcus pneumoniae* and Viridans streptococci. The panel encompasses a total of 16 antimicrobial agents from 12 different classes; however, their inclusion in the template will vary among the organism groups based on the inclusion and exclusion criteria. The CLSI clinical breakpoints can be used to interpret the AST results for most drug/bug combinations, except for β hemolytic streptococci (trimethoprim-sulfamethoxazole, moxifloxacin, teicoplanin and rifampicin), *Streptococcus pneumoniae* (ampicillin and teicoplanin) and Viridans streptococci (teicoplanin); for which EUCAST breakpoints may be used [5].

**Definitions used for drug-resistant bacteria**

The present document upholds the definitions for MDR, XDR and PDR as suggested by the joint ECDC/CDC expert group. Not-susceptibility refers to either a resistant or intermediate or non-susceptible result obtained from antimicrobial susceptibility testing. The bacterial isolate that is not-susceptible to at least one agent in three or more antimicrobial classes should be classified as Multidrug Resistant (MDR) bacteria. The Extensively Drug-Resistant (XDR) bacterial isolate is the one that is not-susceptible to at least one agent in all but two or fewer antimicrobial classes. When the isolate is found not-susceptible to all the antimicrobial agents in all antimicrobial

classes, should be considered as Pandrug Resistant (PDR). The PDR isolates exhibit the highest level of antimicrobial resistance possible, indicating that there are no approved antimicrobial agents that have activity against these strains.



**Figure 1.** Diagram depicting the relationship of MDR, XDR and PDR to each other.

It is also important to understand that the MDR, XDR and PDR are not mutually exclusive terminologies. The XDR is actually a subset of MDR, and the PDR is truly a subcategory of XDR. Thus, a bacterial isolate would have to be MDR in order for it to be further defined as XDR. Similarly, a bacterial isolate that is categorised as PDR will also be characterized as XDR. Figure 1 illustrates that MDR category comprises of six mutually exclusive sub categories: (i) MDR only, (ii) MDR with possible XDR only, (iii) MDR with possible XDR/PDR, (iv) XDR only, (v) XDR with possible PDR only and (vi) PDR only. Similarly, the XDR category includes three distinct sub categories (i) XDR only, (ii) XDR with possible PDR and (iii) PDR only. Appropriate caution should be taken while assigning the isolates into the appropriate drug resistant category. Figure 2 depicts classifying a number of bacterial isolates with different AST patterns into various drug resistant categories as per the proposed definitions for MDR, XDR and PDR.

Isolate	Antibiotic classes									No. of classes			Classification
	1	2	3	4	5	6	7	8	9	R/≥1	R/All	NT	
1	■	■	■	■	■	■	■	■	■	1	1	0	Not MDR
2	■	■	■	■	■	■	■	■	■	2	1	0	MDR only
3	■	■	■	■	■	■	■	■	■	2	2	0	MDR only
4	■	■	■	■	■	■	■	■	■	5	4	0	XDR
5	■	■	■	■	■	■	■	■	■	1	6	0	XDR
6	■	■	■	■	■	■	■	■	■	1	7	0	XDR
7	■	■	■	■	■	■	■	■	■	1	8	0	XDR
8	■	■	■	■	■	■	■	■	■	2	1	6	MDR, Possible XDR only
9	■	■	■	■	■	■	■	■	■	1	4	4	MDR, Possible XDR only
10	■	■	■	■	■	■	■	■	■	1	5	3	MDR, Possible XDR only
11	■	■	■	■	■	■	■	■	■	1	6	2	XDR
12	■	■	■	■	■	■	■	■	■	1	7	1	XDR
13	■	■	■	■	■	■	■	■	■	0	5	4	MDR, Possible XDR/PDR
14	■	■	■	■	■	■	■	■	■	0	6	3	MDR, Possible XDR/PDR
15	■	■	■	■	■	■	■	■	■	0	7	2	XDR, Possible PDR
16	■	■	■	■	■	■	■	■	■	0	8	1	XDR, Possible PDR
17	■	■	■	■	■	■	■	■	■	0	9	0	PDR

■ Indicates that the isolate is susceptible to all agents of that antimicrobial class  
 ■ Indicates that the isolate is susceptible to some agents, but not-susceptible to the other agents of an antimicrobial class  
 ■ Indicates that the isolate is resistant to all the agents of that antimicrobial class  
 ■ Indicates that the isolate is not tested for all the agents of that antimicrobial class  
 ■ Indicates the isolate is susceptible to some agents and not tested for the other agents of an antimicrobial class  
 R/≥1, indicates that the isolate is resistant to ≥1 agents of that antimicrobial class  
 R/All, indicates that the isolate is resistant to all the agents of that antimicrobial class

**Figure 2.** Illustration that depicts classifying a number of bacterial isolates with different AST patterns into various drug resistant categories as per the proposed definitions for MDR, XDR and PDR.

## Discussion

The World Health Organization (WHO) has ranked AMR as one of the top 10 global public health threats facing humanity. Furthermore, the dry clinical pipeline of novel antimicrobials further fuels the emergence and spread of drug-resistant bacteria, leaving behind the clinicians a very limited therapeutic options. There is a large epidemiological disparity in the prevalence of various drug resistant bacteria across the globe. The true burden of drug resistant bacteria viz. MDR, XDR and PDR bacteria may vary between nations, between different provinces of same nation, between community and tertiary care centres, between private and public sector facilities and between different centres of same locality. Therefore, it is essential to monitor the AMR trend in the community through conducting AMR surveillance, which not only helps in providing information about the burden of drug-resistant bacteria at local, national and global level ; but also helps in assessing the effectiveness of AMR preventive efforts. There are several AMR surveillance networks that are currently in operation for collection, analysis and sharing of AMR data; both at a global and national level. 'GLASS' (Global Antimicrobial Resistance Surveillance System), a WHO's initiative of global surveillance network, which aims to support, encourage and facilitate the establishment of national AMR surveillance systems that are capable of monitoring AMR trends and producing reliable and comparable data. In concordance, several national level surveillance systems exist in India. 'AMRSN' (AMR surveillance Network) is an initiative by Indian council of medical research under of Government of India, which is operation since 2013. National Programme on Containment of Anti-Microbial Resistance (AMR) is another surveillance network by NCDC, India.

In spite both these surveillance centres are active since a decade, they could include only few centres to their network (19 centres included in AMRSN, whereas 29 centres included in NCDC's AMR network). The main drawback of these networks is inclusion of a large number of antimicrobials for testing which requires supplemental testing, and therefore need additional man power and fund. The templates provided by ECDC/CDC expert group for the purpose of analysing MDR/XDR/PDR also comprise a long list of antimicrobials, which puts the centres in a difficult position to implement. Most centres routinely perform AST by disk diffusion method and include only a limited antimicrobial disk for testing; which limits their participation to these surveillance networks. Therefore the AMR surveillance networks should look for adjusting their protocols to include a list of antimicrobials that are routinely tested by the laboratories, which obviates the need of additional workforce and budget. This is possible by adapting the methodology from disk diffusion to automated AST systems, which generally use the AST panels comprising of a fixed set of antimicrobial agents.

## Advantages

The present interim proposal provides a guidance for the automated AST user centres to conduct surveillance of MDR, XDR and PDR bacteria. VITEK-2 being the most common automated AST instrument used by clinical, reference or public health microbiology laboratory in India, this proposal provides an opportunity to compare the AST results between these user centres. There are several advantages of surveillance systems based on Automated AST

method over those based on disk diffusion method (i) provision of using a common set of antimicrobials across centres which makes the data comparable, (ii) provision of MIC-based interpretation of AST result, which is more reliable and accurate than that of zone diameter, (iii) provision of using of a common testing protocol by all the centres, as per manufacturer's instruction, (iv) uniformity of the technical expertise, as provided by the manufacturer, thus obviates the need of additional training.

## Challenges while implementing the proposal

While implementing the proposed interim guideline to the AST results obtained from the automated instrument, user centres may face few challenges. Therefore, the centres must ensure that they fulfil certain criteria to obviate the challenges.

- **Users of same automated equipment:** This proposal is developed only for the VITEK-2 user centres. The centres which use other automated equipment such as BD Phoenix and MicroScan should prepare their own templates by following the same principles used in this document.
- **Use of the same AST panel:** It is important that the centres must use the same automated AST panels routinely for performing AST, which will make their AST data comparable.
- **Use of the same breakpoint:** Centres must adapt to the same interpretation criteria (for e.g., CLSI, EUCAST, FDA, other in decreasing order of preference).
- **Uniformity of the testing method:** It is also worth mentioning that the testing method must be uniform across all centres with appropriate quality standards. This can be made possible through constant training, provided by the manufacturer.
- **Validation by AES:** The AST results that are flagged 'consistent' should be included for analysis. Those 'flagged consistent with correction' may be included for analysis only after manual confirmation by the laboratory; whereas those which flagged 'inconsistent' must be repeated by automated AST system after confirming the purity of the isolate.
- **Inclusion of suppressed antibiotic data:** It is recommended that the antimicrobial agents that might be suppressed on patient report (due to cascade reporting practice of the laboratory) should be added for analysis.
- **Reconfirmation of suspicious AST results:** It is important that the centres should reconfirm the suspicious AST results (e.g. linezolid resistance in *S. aureus*) by another recommended method before including for analysis.
- **Exclusion of critical care AST panel:** The critical care AST panel is used by the centres as a supplemental panel only for the isolates that are found resistant to the primary AST panels. Therefore, such panels should be excluded from analysis.
- **Limiting the proportion of not tested antimicrobials:** If the AST results are not obtained for some antimicrobial agent due to partial termination or some other cause, the centres should repeat the test to obtain the result. Although it is no always practicable to obtain the AST result for all the antimicrobials listed in templates for all the organisms all the time. However, the centres must exercise utmost care to limit the proportion of 'not tested' antimicrobials. Otherwise it will create misinterpretation of the exact category of the drug resistant bacteria; for e.g., an

isolate with 'MDR only' category may be interpreted wrongly as 'MDR with possible XDR'.

- **Divergent resistance profiles of MDR strains:** When using 'MDR' as a parameter to characterize drug resistant bacteria of public health significance, it is of note to understand an important limitation in the definition of MDR used in this document. Bacterial isolates with diverse resistance profiles (resistant to different set of  $\geq 3$  classes) may still fit to the definition of MDR (i.e. not-susceptible to  $\geq 1$  agent of  $\geq 3$  classes). Further analysing the resistance profile of these MDR isolates is beyond the scope of these definitions.
- **Minimum number of isolates:** It is also important to note that minimum 30 number of isolates of an organism per centre must be included for analysis in order to achieve a statistically significant comparison between their MDR, XDR/PDR data.
- **Dealing with site specific antimicrobials:** While determining the MDR/XDR/PDR category of an organism from a particular site, the site specific antimicrobials (e.g nitrofurantoin for urinary isolates) must be added for analysis.
- **Updating the templates:** The templates are developed based on the current automated AST panels available for use. The templates need to be revised on a regular basis whenever the AST panels are revised by the manufacturer.

## Conclusion

In the era of increased AMR, it is important that the healthcare facilities should conduct surveillance to know the true estimate of the drug resistant bacteria prevalent in their centres. However, it is impractical to obtain the MDR/XDR/PDR data for those centres that follow disk diffusion test for AST, because of the differences in the antimicrobial agents that are used for testing. This interim guideline provides a direction to the automated AST user centres for analysing their AMR data. Applying the templates given in this proposal, the facilities can determine their MDR/XDR/PDR data which can be comparable between various automated AST user centres across the globe. The use of automated systems (especially VITEK-2 in India) for performing AST has been increasing in the recent past and is expected to further expand in future. Furthermore, there is no need for any additional manpower or budget, as the analysis of drug resistant bacteria is performed based on the routine AST data. Therefore, a large number of user centres can contribute their AMR data, which can be collated to give a true picture of the current

burden of MDR/XDR/PDR in the World. This information is essential for developing empirical antimicrobial therapy for diverse epidemiological settings.

## Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

## Conflicts of Interest

The authors have no conflicts of interest to declare that are relevant to the content of this article.

## Ethics Approval

Not required.

## References

1. Ryu, Sukhyun, Cowling Benjamin J, Wu Peng, and Olesen Scott, et al. "Case-Based Surveillance of Antimicrobial Resistance with Full Susceptibility Profiles." *JAC-Antimicrob Resis* 1 (2019): dlz070. Magiorakos, A P, Srinivasan A, Carey R B, and Carmeli Y, et al. "Multidrug-Resistant, Extensively Drug-Resistant and Pandrug-Resistant Bacteria: An International Expert Proposal for Interim Standard Definitions for Acquired Resistance." *Clin Microbiol Infect* 18 (2012): 268-281.
2. Falagas, Matthew E, Koletsis Patra K, and Bliziotis Ioannis A. "The Diversity of Definitions of Multidrug-Resistant (MDR) and Pandrug-Resistant (PDR) *Acinetobacter Baumannii* and *Pseudomonas Aeruginosa*." *J Med Microbiol* 55 (2006): 1619-1629.
3. van Belkum, Alex, Bachmann Till T, Lüdke Gerd, and Lisby Jan Gorm, et al. "Developmental Roadmap for Antimicrobial Susceptibility Testing Systems." *Natu Rev Microbiol* 17 (2019): 51-62.
4. Felmingham, David, and Brown Derek FJ. "Instrumentation in Antimicrobial Susceptibility Testing." *J Antimicrob Chemother* 48 (2001): 81-85.

**How to cite this article:** Sankar, Sastry Apurba, Ketan Priyadarshi, Deepashree Rajashekar, and Sumit Rai. "Interim Proposal for Surveillance of Multidrug-Resistant, Extensively Drug-Resistant and Pandrug-Resistant Bacteria for the Automated AST User Centres ." *J Antimicro Agent* 7 (2021) : 251