

# New CLSI and EUCAST harmonized procedure for selecting optimal disk contents for disk diffusion testing

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## Background

Standardized methods for performing disk diffusion susceptibility testing, which has been used worldwide for decades, are published by both CLSI and EUCAST. However, disk contents recommended by either CLSI or EUCAST for a specific antimicrobial agent sometimes differ. To overcome this problem, CLSI and EUCAST in 2018 assembled a joint working group to develop a harmonized procedure for selecting optimal disk contents.

## Methods

Publications of relevance were consulted and data from previous disk content selection studies were examined. Knowledge and experience in disk diffusion and selecting disk content of working group members were relied upon to develop the procedure.

## Results

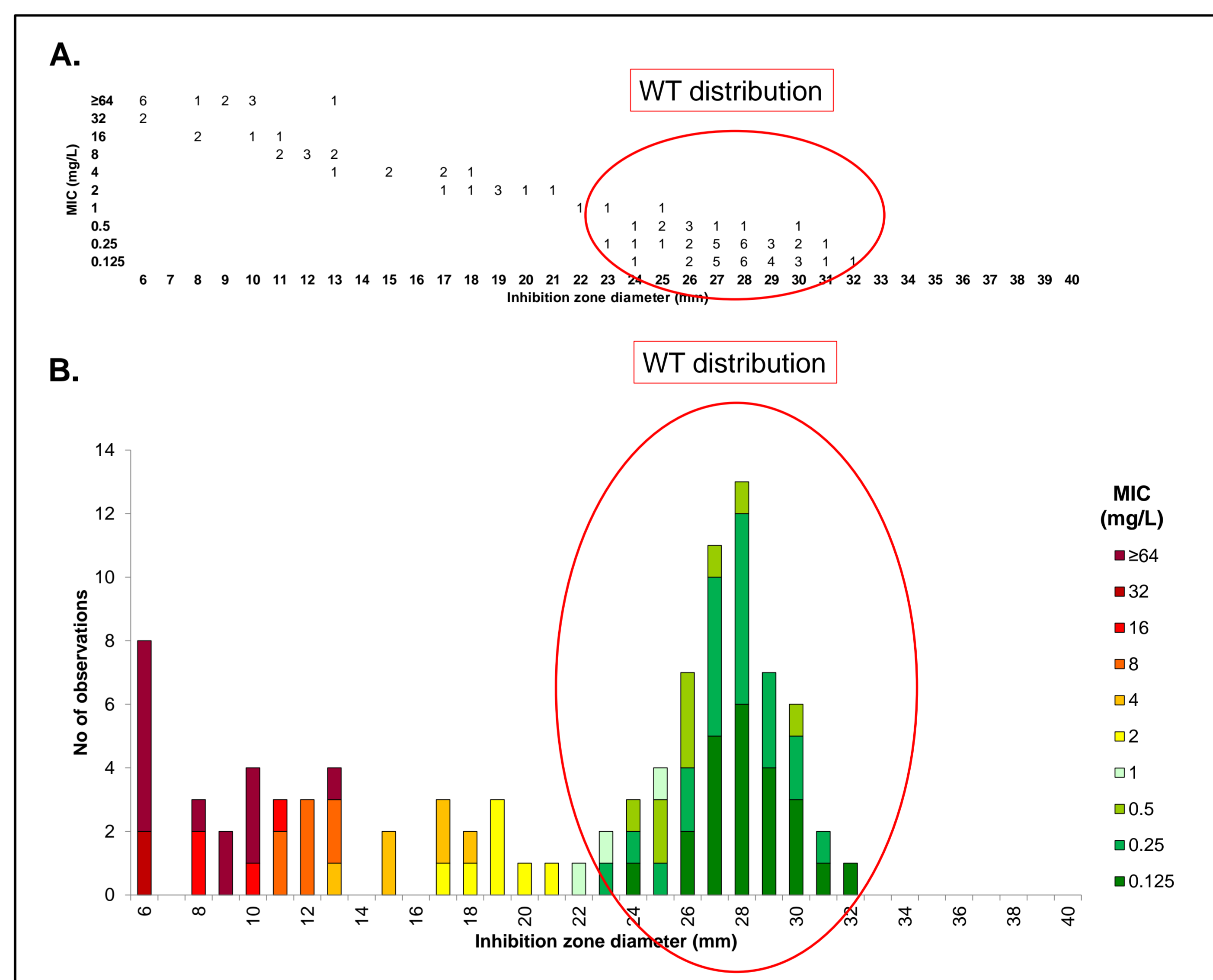
The agreed selection criteria for disks containing a single antimicrobial agent are:

### When feasible, studies are performed to achieve:

- Reproducible inhibition zone diameters when testing QC strains and clinical isolates
- A single disk content (potency) that can be used for all relevant species (target organisms)
- A general discriminatory power of 2- to 3-mm increase in zone diameters with each log<sub>2</sub> decrease in MIC for non-wild type (NWT) isolates
- Inhibition zone diameters between 15 and 35 mm (ideally not above 30 mm) for WT isolates of relevant species (target organisms)
- Optimal separation between WT and NWT isolates (when MIC clinical breakpoints are not yet defined), if NWT isolates exist
- Optimal separation between NWT isolates with different MICs, irrespective of resistance mechanisms

Testing is performed in two phases. **In phase 1**, 10 different disk contents ranging from very low to very high contents (e.g. 0.1-100 µg) are typically tested against at least four isolates per relevant target species. The 2-4 disk contents from phase 1 which best meet the selection criteria listed above are used for phase 2.

**In phase 2**, both wild-type isolates and isolates with elevated MICs for the agent are selected. The number of isolates for phase 2 is larger than for phase 1 (typically 30 per target species and 60 per organism group) and testing is performed with two lots of disks and media from two manufacturers. Data for each disk potency and species/organism group are presented as scattergrams and MIC-zone diameter correlation histograms (examples in **Figure 1**) and examined for conformity with the listed selection criteria.



**Figure 1. MIC-zone diameter correlations for a single disk potency as A) Scattergram and B) Zone diameter histogram with corresponding MICs as coloured bars (with green colours corresponding to MICs for WT isolates). Figures A and B represent the same dataset.**

## Conclusions

The harmonized CLSI and EUCAST procedure for selecting optimal disk contents for disk diffusion testing is now available online (CLSI: M23S, EUCAST: SOP 11.0). Recognition of a single disk content by both CLSI and EUCAST for each antimicrobial agent is beneficial to pharmaceutical companies, manufacturers of antimicrobial disks and laboratories performing disk diffusion testing.

Access to the procedure on CLSI and EUCAST websites

CLSI (M23S): <https://www.clsi.org/standards/products/microbiology/documents/m23s/>

EUCAST (SOP 11.0): <https://www.eucast.org/eucastsops>