

Get to know your options: The importance of conventional antimicrobial susceptibility testing (AST) in a clinical laboratory

What is conventional AST?

Conventional antimicrobial susceptibility testing (AST) is a laboratory procedure used to determine the effectiveness of antibiotics against isolated bacteria and fungi from clinical samples. Conventional AST methods include disc diffusion (Kirby-Bauer), broth dilution, and agar dilution. AST is essential for guiding appropriate antibiotic therapy and combating antibiotic resistance.

Disc diffusion is one of the most simple and widely used methods for susceptibility testing. Disc testing is an extremely flexible technique, with a wide variety of antibiotics and concentrations available, which can easily be interchanged as required.

The disc diffusion test not only categorizes resistant, intermediate and susceptible organisms through quantitative results, but also provides a visual indication of:

- Inoculum level
- Presence of contamination
- Antagonism and synergy between adjacent antibiotics
- β-lactamase activity

Although disc diffusion is a simple test to perform, to obtain accurate and reproducible results attention to detail and care must be taken, since relatively small changes in operating procedure can affect the results obtained.

How does conventional AST work?

- **1. Isolation**: Bacteria or fungi are cultured from a clinical sample.
- **2. Inoculum preparation**: Bacterial or fungal colonies are suspended in saline or broth to a standardized concentration.
- **3. Inoculation**: The suspension is spread onto an agar plate.
- 4. Application of antibiotics: Antibiotic-impregnated discs are placed on the inoculated agar surface.
- 5. Incubation: The plates are incubated to allow antibiotic diffusion and bacterial growth.
- 6. Result interpretation: The zones of inhibition around the discs are measured and compared to standard criteria to classify the bacteria as susceptible, intermediate, or resistant.

In a disc diffusion test, bacterial and fungal isolates are tested for their susceptibility to different antibiotics. A clear visible large ring of no growth around an antibiotic disc is called a zone of inhibition. This inhibition zone shows that the antibiotic has impeded the growth of the organism indicating that the organism is susceptible. Bacterial or fungal growth with no zone of inhibition indicates the organism is not affected by the antibiotic and is resistant. CLSI (Clinical and Laboratory Standards Institute) and EUCAST (European Committee on Antimicrobial Susceptibility Testing) are organizations that provide guidelines and standards for antimicrobial susceptibility testing. In order to promote reproducibility and comparability of results between laboratories these standard methods are followed. These standards provide guidelines for antimicrobial susceptibility testing and establish breakpoints using a comprehensive approach that includes clinical, pharmacokinetic/pharmacodynamic and microbiological data.

How was conventional AST developed?

- The concept of testing the effectiveness of antimicrobial agents can be traced back to the early 1900s, following the discovery of penicillin by Alexander Fleming in 1928. Initial methods were not standardized and involved simple observations of bacterial growth inhibition in the presence of antimicrobial agents.
- During the 1940s, with the mass production and widespread use of antibiotics, the need for standardized testing methods became apparent.
- The introduction of the disk diffusion method provided a more systematic approach. This method was further refined and standardized over the next few decades.
- In the 1950s the Kirby-Bauer disk diffusion method, named after scientists William Kirby and Alfred Bauer, was developed and became widely accepted. This method involves measuring the zone of inhibition around antibiotic disks to determine bacterial susceptibility.
- The Kirby-Bauer method was officially standardized by various organizations and became a cornerstone of antimicrobial susceptibility testing.
- This was followed by further advancements included the development of broth dilution methods (both macro and micro), which allowed for the determination of minimum inhibitory concentrations (MICs) – the lowest concentration of an antibiotic that inhibits visible organism growth.
- Organizations such as the CLSI and EUCAST have played crucial roles in providing standardized guidelines and protocols for AST.
- Modern AST methods supplement conventional AST and include automated systems, molecular techniques, and advanced technologies to rapidly and accurately determine microbial susceptibility to a wide range of antimicrobial agents.

Why is AST so important?

AST is critically important for several reasons:

1. Guiding effective treatment

- Personalized therapy: Helps determine the most effective antibiotics for treating a specific bacterial or fungal infection in an individual patient.
- Avoiding ineffective drugs: Helps to avoid the use of antibiotics that microorganisms are resistant to, supporting better clinical outcomes.

2. Combating antibiotic resistance

- **Surveillance**: Tracks patterns of antibiotic resistance, which is vital for public health monitoring and response.
- Informed prescribing: Helps to reduce the misuse and overuse of antibiotics, which are major drivers of resistance.

3. Guidance for clinical decision-making

- Empirical therapy adjustment: Supports clinicians to adjust initial empirical antibiotic therapy based on susceptibility results, helping to improve treatment accuracy.
- Support for severe infections: Guide clinicians in managing serious infections where timely and appropriate antibiotic therapy is important.

4. Public health and epidemiology

- Data collection: Provides data for local, regional, and global surveillance of resistance trends.
- **Policy development**: Informs guidelines and policies for antibiotic use in healthcare settings.

5. Supporting infection control

- **Outbreak management**: Identifies resistant strains during outbreaks, aiding in containment and control measures.
- Hospital infection control: Helps in developing and implementing infection control protocols to prevent the spread of resistant organisms.

6. Research and development

- New antibiotics: Assists in the development and testing of new antibiotics by providing data on microorganism resistance patterns.
- Resistance mechanisms: Facilitates research into the mechanisms of resistance, which can lead to new treatment strategies.

AST is a cornerstone of clinical microbiology and infectious disease management, playing a crucial role in guiding clinicians in determining effective treatment, combating antibiotic resistance, and supporting public health initiatives.

Thermo Scientific AST solutions

Thermo Scientific[™] Oxoid[™] Antimicrobial Susceptibility Test (AST) Discs

Fight antimicrobial resistance with the simplicity and flexibility of disc diffusion using Thermo Scientific Oxoid AST Discs. Available in a variety of common and specialized compounds, and a full range of concentrations, Oxoid AST Discs allow high quality results with many meeting the applicable CLSI or EUCAST requirements.

Oxoid AST Discs are backed by over 50 years of experience in antimicrobial susceptibility disc development and production. By maintaining close relationships with many pharmaceutical companies this enables the introduction of new discs to the market in a timely manner when new compounds become available.

To ensure maximum accuracy and reproducibility, Oxoid AST discs are manufactured in accordance with appropriate quality standards, including ISO 9001:2015, ISO 13485:2016 and FDA 21 CFR part 820, and undergo comprehensive quality control testing. For clinical applications, Oxoid discs intended for the EU market are CE-marked under the IVD directive, 98/79/EC or IVD Regulation (EU) 2017/746. In addition:

- Test methods and specifications (zone size) adhere to the applicable standard body guidelines with disc performance being the principal quality parameter
- Batch variation, moisture and concentration testing are also performed utilizing an antibiotic concentration specification of 90% to 125%* of the stated concentration



Thermo Scientific[™] Oxoid[™] and Remel[™] culture media

The Thermo Scientific culture media portfolio offers a wide variety of options for greater testing flexibility, including dehydrated and prepared plated culture media, plus tubed broths and agars in a variety of sizes and fill volumes. This range includes core culture media for AST - including Mueller Hinton Agar and Broth, and non-selective, nutrient rich blood agars to ensure reliable AST results.

Backed up by over 100 years of experience, Thermo Scientific culture media are manufactured with rigorous quality standards and control of every step of the manufacturing process from raw materials to finished product, providing a trusted source of culture media for laboratories around the world.

Thermo Scientific[™] Sensititre[™] AST System

The Sensititre AST System is a broth microdilution testing platform with scalable testing options from conventional broth microdilution plate set up to semi-automated to fully automated workflows supported by instrumentation and result interpretation software with LIS connectivity.

The system delivers accurate MIC results, and supports a wide range of pathogens and antimicrobials, ensuring comprehensive testing capabilities to meet the specific needs of different healthcare settings and a range of products guiding clinicians to make informed decisions to combat infections effectively.



Antimicrobial susceptibility testing is crucial for guiding appropriate antibiotic therapy and helping to combat antibiotic resistance. While it requires careful technique and standardized procedures, it remains a cornerstone in clinical microbiology labs.

For more information on how to find solutions perfectly matched for your AST program, please contact your local Thermo Fisher Scientific Microbiology representative or visit **thermofisher.com/AST**

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