

Disinfectants, Antimicrobial Agent Susceptibility Testing, and Resistance

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Disinfection, antimicrobial susceptibility testing, and antimicrobial resistance are related through a common objective—controlling microorganisms—but they answer different questions. Disinfection asks how microorganisms can be reduced or inactivated on surfaces, equipment, or other nonliving materials. Antimicrobial susceptibility testing (AST) asks whether a clinical isolate is likely to respond to a therapeutic antimicrobial under standardized conditions. Antimicrobial resistance explains why organisms can survive drug exposure that would ordinarily inhibit or kill susceptible strains. Confusing these domains leads to practical errors, such as assuming that a disinfectant active on a bench surface can be used safely on living tissue or interpreting an inhibition zone without reference to current clinical breakpoints.

Current laboratory and infection-control guidance makes standardization central. The CDC continues to recommend meticulous cleaning before high-level disinfection or sterilization because residual organic and inorganic material can interfere with antimicrobial action (CDC, 2023a). For AST, the Clinical and Laboratory Standards Institute's 2026 M100, 36th Edition, provides updated breakpoints and quality-control criteria for use with standardized disk-diffusion and dilution methods, while EUCAST's 2026 breakpoint tables similarly connect measured MICs or zone diameters with therapeutic interpretation (CLSI, 2026; EUCAST, 2026a). These systems show why reliable microbial control depends on method, concentration, contact time, organism, and clinical context rather than on the label "strong antimicrobial."

Disinfection Begins With Risk Classification and Cleaning

Disinfection is the use of chemical or physical processes to inactivate microorganisms on inanimate objects to a level appropriate for their intended use. It differs from antisepsis, which applies antimicrobial agents to living tissue, and from sterilization, which is intended to eliminate all forms of viable microbial life, including bacterial spores, under validated conditions. The distinction matters because patient-care items present different risks. The Spaulding framework classifies devices as critical, semicritical, or noncritical according to how they contact the patient, and that classification helps determine whether sterilization, high-level disinfection, or lower-level disinfection is required (CDC, 2023b).

Cleaning comes before disinfection or sterilization for many reusable medical items. Blood, proteins, salts, tissue, feces, biofilm, and other material can shield microorganisms or chemically reduce the activity of disinfectants. The CDC therefore recommends prompt and thorough removal of visible organic and inorganic residue before high-level disinfection or sterilization (CDC, 2023a). A chemical agent cannot reliably disinfect a surface that it cannot reach.

Disinfectant activity depends on several interacting variables. Concentration must fall within the validated range; underdilution may reduce efficacy, while excessive concentration can increase toxicity, damage equipment, or fail to improve antimicrobial performance. Contact time is the period during which the surface remains exposed to the product at the required concentration. A spray that is wiped away immediately may not achieve the label claim. Temperature and pH can alter chemical stability and microbial susceptibility. Organic matter can reduce activity, while cracks, lumens, porous materials, and biofilms make organisms harder to reach. Microbial load also matters because a larger starting population requires greater log reduction to achieve the same final level of contamination.

Different organisms have different intrinsic resistance. Bacterial spores are generally more difficult to inactivate than vegetative bacteria, while mycobacteria and many nonenveloped viruses can also require stronger processes than enveloped viruses. Prions require specialized procedures outside ordinary disinfection assumptions. The idea that all Gram-positive bacteria are “easy to kill” and all Gram-negative bacteria are “hard to kill” is therefore too crude for infection-control decisions. Product-specific efficacy claims and validated reprocessing methods are more reliable than one universal hierarchy.

Regulatory status is also important. In the United States, many environmental surface disinfectants are regulated by EPA, whereas FDA regulates certain liquid chemical sterilants and high-level disinfectants intended for reusable medical devices. FDA-cleared products specify validated contact conditions, concentrations, temperatures, reuse periods, and device applications; those conditions are not interchangeable among products (FDA, 2026). Healthcare workers should follow both the disinfectant label and the device manufacturer’s reprocessing instructions.

Antimicrobial Susceptibility Testing Is a Measurement System, Not a Visual Guess

AST determines how a bacterial or fungal isolate responds to therapeutic [antimicrobial agents](#) under standardized laboratory conditions. The result can support drug selection, antimicrobial stewardship, resistance surveillance, and infection-control investigation. The central measurements are usually a minimum inhibitory concentration (MIC) or an inhibition-zone diameter. Neither measurement has clinical meaning by itself; it must be interpreted using a current breakpoint system.

AST component	Why it matters
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Pure identified isolate	Breakpoints and intrinsic-resistance rules are organism-specific; mixed cultures cannot usually be interpreted as one susceptibility result.
Standardized inoculum	Too many organisms can make an isolate appear more resistant; too few can make it appear more susceptible.
Validated medium and incubation	pH, cations, agar depth, atmosphere, temperature, and incubation duration influence antimicrobial activity and growth.
MIC or zone diameter	Provides the measured phenotype used for interpretation.
Current breakpoint	Connects the measurement with dosage, organism, infection site, PK/PD, and clinical evidence.
Quality-control strain	Confirms that media, reagents, disks, instruments, and technique are functioning within expected limits.

Disk diffusion uses a standardized microbial suspension spread across agar, followed by antimicrobial disks and controlled incubation. The diameter of each inhibition zone is measured and compared with current interpretive criteria. A larger zone does not mean one antimicrobial is universally “stronger” than another because each drug has different diffusion characteristics and breakpoints. A zone indicates inhibition under the test conditions, not necessarily bacterial killing.

Broth dilution determines the MIC, the lowest concentration preventing visible growth under standardized conditions. Reference broth microdilution remains foundational for bacterial AST, and commercial automated systems are calibrated against reference methods. Gradient diffusion can also estimate an MIC on agar. CLSI’s 2026 M100 explicitly depends on methods standardized in M02, M07, and M11, emphasizing that breakpoint tables are valid only when the underlying methodology is followed correctly (CLSI, 2026).

EUCAST uses the categories S, I, and R with definitions linked to exposure. S means susceptible at a standard dosing regimen. I means susceptible when exposure is increased through dose, dosing interval, infusion, distribution, or concentration at the infection site. R means a high likelihood of therapeutic failure even with increased exposure (EUCAST, 2026b). The I category should therefore not be incorrectly grouped with resistance. EUCAST’s current bacterial breakpoint table is version 16.1 for the second half of 2026, illustrating why laboratories cannot rely indefinitely on old interpretive charts (EUCAST, 2026a).

Controls are essential. A growth control confirms that the organism can grow under the conditions of the test, while a sterility control confirms that uninoculated media are not contaminated. Quality-

control strains with established acceptable ranges evaluate the entire testing system. If the growth control fails, an apparently “susceptible” result may simply reflect a nonviable inoculum. If a control strain falls outside its expected range, patient results should not be trusted until the cause is investigated.

Resistance Is Biological, but the Laboratory Definition Is Operational

Microorganisms can resist therapeutic agents through several mechanisms: drug-inactivating enzymes, alteration of the antimicrobial target, reduced permeability, active efflux, metabolic bypass, target protection, biofilm-associated tolerance, and acquisition of resistance genes through plasmids, transposons, integrons, or bacteriophages. Mutation followed by selection can also produce resistance. Common examples include beta-lactamases, altered penicillin-binding proteins, ribosomal modification, and mutations affecting fluoroquinolone targets.

Clinical resistance, however, is not defined simply as “the organism survives the drug.” Breakpoints integrate microbiology with pharmacokinetics, pharmacodynamics, dosage, infection site, organism-specific MIC distributions, resistance mechanisms, and clinical outcome evidence. EUCAST explains that breakpoint setting is a scientific process requiring all of these forms of evidence rather than one mathematical threshold applied to every organism–drug pair (EUCAST, 2026c).

This is why breakpoints can change even when the organism has not. New dosage regimens, pharmacodynamic data, resistance mechanisms, or clinical outcomes may justify reinterpretation of the same MIC. Laboratories should use the current standard and document which system and version they apply. Mixing CLSI and EUCAST breakpoints within one report without a defined policy can produce inconsistent clinical interpretations.

Routine AST also has limitations. Planktonic organisms tested in standardized media may behave differently from organisms embedded in a biofilm on a catheter, prosthetic device, or damaged tissue. Heteroresistance can leave small subpopulations with higher resistance than the dominant culture. Molecular tests can detect particular resistance genes rapidly but do not identify every possible mechanism and do not always predict expression perfectly. Phenotypic and genotypic methods should therefore be understood as complementary rather than interchangeable.

Disinfectant Tolerance and Antibiotic Resistance Should Not Be Conflated

Disinfectants and therapeutic antibiotics differ greatly in purpose and exposure. Surface disinfectants are generally used at much higher concentrations and are intended for nonliving environments, while

antibiotics must reach effective concentrations in patients without causing unacceptable toxicity. The mechanisms of reduced susceptibility can nevertheless overlap. Efflux pumps, permeability changes, biofilm formation, stress responses, or genetic linkage between resistance determinants can create concern that repeated sublethal biocide exposure might co-select organisms with reduced antibiotic susceptibility.

The evidence does not justify avoiding necessary disinfection. Poor infection prevention would increase transmission and antibiotic use, worsening antimicrobial resistance. The practical lesson is to avoid sublethal and inappropriate use: prepare disinfectants correctly, clean first, maintain the full validated contact time, avoid contamination of dispensers, and do not use antimicrobial chemicals where ordinary cleaning is sufficient.

Antibiotic stewardship is equally important. WHO's 2025 global surveillance report analyzed more than 23 million bacteriologically confirmed infections and found substantial antibiotic resistance across common pathogens and drug combinations worldwide (WHO, 2025a). The same year, WHO reported major gaps in the availability and access to rapid pathogen identification and susceptibility diagnostics, emphasizing the need for both phenotypic AST and newer diagnostic approaches (WHO, 2025b). Laboratory quality therefore contributes directly to stewardship: an inaccurate resistant result can drive unnecessary broad-spectrum therapy, while a false susceptible result can delay effective treatment.

Quality Assurance Connects Infection Control With Patient Care

Both disinfection and AST are vulnerable to errors that appear small but change the final conclusion. A disinfectant may fail because the surface was not cleaned, the concentration was incorrect, or contact time was too short. An AST result may fail because the inoculum was too heavy, the agar was too deep, incubation conditions were wrong, disks were degraded, or outdated breakpoints were used. Quality assurance makes these processes reproducible.

Healthcare facilities should therefore define reprocessing procedures according to device risk and manufacturer instructions, train staff, monitor concentrations or reuse limits where required, and investigate failures. Clinical microbiology laboratories should use validated methods, quality-control organisms, competency assessment, current CLSI or EUCAST criteria, and procedures for confirming unusual resistance phenotypes.

The larger lesson is that antimicrobial control depends on matching a method to a purpose. Disinfection reduces environmental or device contamination; sterilization provides a higher level of assurance for critical items; susceptibility testing predicts the likely activity of therapeutic antimicrobials; and resistance mechanisms explain why organisms may escape those interventions. Reliable practice does not come from using the strongest available chemical. It comes from

standardized preparation, measurement, interpretation, and quality control.

References

Centers for Disease Control and Prevention. (2023a). *Cleaning: Guideline for Disinfection and Sterilization in Healthcare Facilities*.

Centers for Disease Control and Prevention. (2023b). *A Rational Approach to Disinfection and Sterilization*.

Clinical and Laboratory Standards Institute. (2026). *Performance Standards for Antimicrobial Susceptibility Testing* (M100, 36th ed.).

European Committee on Antimicrobial Susceptibility Testing. (2026a). *Clinical Breakpoint Tables for Bacteria, Version 16.1*.

European Committee on Antimicrobial Susceptibility Testing. (2026b). *Definitions of S, I and R*.

European Committee on Antimicrobial Susceptibility Testing. (2026c). *Setting Clinical Breakpoints*.

U.S. Food and Drug Administration. (2026). *FDA-Cleared Sterilants and High-Level Disinfectants for Reusable Medical and Dental Devices*.

World Health Organization. (2025a). *Global Antibiotic Resistance Surveillance Report 2025*.

World Health Organization. (2025b). *Landscape Analysis of Commercially Available and Pipeline In Vitro Diagnostics for Bacterial Priority Pathogens*.