

Use of gradient tests for determination of MICs by BSAC methodology

The gradient test (Etest, AB Biodisk; MICE, Oxoid) is a method for determining minimum inhibitory concentrations (MICs) and is a practical alternative to conventional agar or broth dilution MIC determinations. In diagnostic laboratories the main uses of the method are for confirming resistance (particularly when equivocal), for testing 'difficult' organisms, and in situations where an MIC is needed, e.g. endocarditis.

Gradient tests are calibrated to give performance substantially equivalent to United States CLSI reference methods. The BSAC reference methods differ from CLSI, particularly in the media used and it would be more convenient to use similar conditions for gradient tests as used in routine tests by BSAC methods. With some modifications gradient tests can be used with conditions recommended for BSAC methods, in which case the interpretative criteria and MICs for control strains should be based on the published recommendations¹ or data on the BSAC web site.

Guidance for the use of gradient tests by BSAC methodology:

The test conditions recommended in the BSAC reference method should be used, with the following exceptions.

1. An inoculum equivalent to a 0.5 McFarland standard without further dilution should be used for MIC determinations including the detection of reduced susceptibility to teicoplanin in CNS.
2. In order to optimise the detection of heterogeneous glycopeptide resistance in *Staphylococcus aureus* Brain Heart Infusion agar (BHIA) should be used with an inoculum equivalent to a 2 McFarland standard. False positives are not uncommon by this method and positives should be confirmed with a population analysis method. Non-heterogeneous low-level glycopeptide resistance is rare and is indicated in tests on BHIA or Mueller-Hinton agar (MHA) but not on Iso-Sensitest agar (ISA).
3. Carbapenem testing on ISA may fail to indicate the presence of metallo- β -lactamases and testing on MHA is more reliable. However, the Etest metallo- β -lactamase test gives some false-positive results so positive results should be confirmed by a reference laboratory.
4. In reading gradient tests, zones of inhibition should be closely examined to detect small colonies within the zones. In particular, cell lysis of pneumococci occurs more rapidly on ISA + blood than on MHA + blood, which may result in less distinct end-points. Tails of zones must be closely examined to avoid reporting falsely low MICs.

Reference

1. Antimicrobial Susceptibility Testing: BSAC Working Party Report. JAC Supplement S1 to volume **48** July 2001.