

Metronidazole MT 5 mcg

SD020

Metronidazole MT 5 mcg discs are used for antimicrobial susceptibility testing of anaerobic organisms.

Composition

*Ingredients	Concentration
Metronidazole	5 mcg/disc

Susceptibility Test Procedure:

1. Prepare Brucella agar with Hemin and Vitamin K1 supplement with 5 % v/v sterile defibrinated sheep blood (M1039).
2. Inoculum is prepared by picking five or more well isolated colonies of similar morphology from 24 to 48 hours old culture grown on Brucella Blood agar.
3. Colonies are suspended in 5 ml of sterile Brucella Broth or other clear broth compare the inoculum turbidity with that of standard 0.5 McFarland (prepared by mixing 0.5 ml of 1.175%barium chloride and 99.5 ml of 0.36N sulfuric acid). Dilute the inoculum or incubate further as necessary to attain comparative turbidity. Alternatively, the inoculum can be standardized by other appropriate optical method (0.08 - 0.13 OD turbid suspension at 625 nm)
4. Dip a sterile non-toxic cotton swab in the inoculum and swab the prepared culture.
5. Aseptically incorporate the discs in the medium.
6. Incubate immediately at $35 \pm 2^{\circ}\text{C}$ under anaerobic conditions and examine after 24 hours or longer, if necessary.
7. Measure the zones showing complete inhibition and record the diameters of the zones to the nearest millimeter using a calibrated instrument like zone scales (PW096/PW297)

Principle:

Brucella Agar Base w/ Hemin and Vitamin K1 is a modified and highly enriched medium, which can be used for the isolation of Brucella and other anaerobic bacteria. The medium contain tryptone, peptone and yeast extract serves as sources of carbon, nitrogen, long chain amino acids and essential growth nutrients including B-complex vitamins. Dextrose serves as a source of energy. Addition of blood provides nutrients and helps to differentiate hemolytic organisms. Presence of hemin and Vitamin K1 supports growth of other fastidious bacteria like Bacteroides species and gram-positive spore bearers like Clostridium species.

Antimicrobial susceptibility tests are either quantitative or qualitative. Disc diffusion test is a qualitative test method. The National Committee for Clinical Laboratory Standards (NCCLS), now known as Clinical Laboratory Standards Institute (CLSI) has published comprehensive documents regarding the disc diffusion systems. The agar disc diffusion test is the most convenient and widely used method for routine antimicrobial susceptibility testing. In subsequent and current practice, antimicrobial impregnated paper discs are applied onto the agar surface. Based on the Bauer-Kirby Method, standardized reference procedures for the disc systems were published by WHO and FDA and are periodically updated by the CLSI (formerly NCCLS)(2). For any antimicrobial testing, Quality control or clinical testing, the method to be followed is the same as mentioned above. However few precautions are to be maintained while handling of the Sensitivity discs,

- On receipt the discs are to be immediately stored at the recommended temperature.
- Medium preparation, Inoculum preparation and incubation to be done as specified.

Interpretation:

Interpret the results on the basis of growth obtained when compared to positive control tubes. Lesser the growth more effective is the antibiotic concentration.

Quality Control:

Appearance: Filter paper discs of 6mm diameter with printed "MT 5" on centre of each side of the disc.
Cultural response: Average diameter of zone of inhibition observed on Brucella agar with Hemin and Vitamin K1 supplement with 5 % v/v sterile defibrinated sheep blood (M1039). Incubated anaerobically at 35°C for 24 - 48 hours.

Organisms (ATCC)	Std. zone of diameter (mm)
<i>Cl. perfringens</i> (ATCC 12924)	26-34
<i>Cl. perfringens</i> (ATCC 13124)	26-34
<i>B fragilis</i> (ATCC 25285)	38-44

Storage and Shelf-life:

Discs should always be stored at -20°C to +8°C under dry conditions, along with the dessicator provided in individual pack. Use before expiry date on the label.

References:

1. Bauer, Kirby, Sherris and Turck, 1966, Am. J. Clin. Path., 45: 493
2. Performance standards of Antimicrobial Disc Susceptibility Tests, M100S, 32nd Ed., CLSI Vol. 42 No.2, Feb-2022.
3. EUCAST, Breakpoint tables for interpretation of MIC's & zone diameters, version 12.0, valid from 01.01.2022.

Note :

Use following media to carry out susceptibility test

For rapidly growing aerobic organisms : Mueller Hinton Agar (M173/M1084)

For *Haemophilus* spp : Haemophilus Test Agar (M1259 + FD117)

For *S.pneumoniae* : Muller Hinton Agar supplemented with 5% Sheep Blood

For *Neisseria* spp : G.C.Agar +1% defined growth supplement (M434 + FD025)

* Not for Medicinal Use



In vitro diagnostic medical device



CE Marking

On receipt store at -20°C



Storage temperature



Do not use if package is damaged



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Disclaimer :

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