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Section: Technical Manual	Subject Title: X And V Disks for Identification of Haemophilus	
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X AND V DISKS FOR IDENTIFICATION OF HAEMOPHILUS

Principle

Haemophilus spp. have different requirements for the growth factors X (hemin) and V (NAD). These requirements are determined based on the presence or absence of growth around disks impregnated with V, X and XV factors.

Reagents

1. Bacto Differentiation Disks
 - BV NAD and 125 units/ml bacitracin
 - BX hemin and 125 units/ml bacitracin
 - BVX NAD, hemin 125 units/ml bacitracin
 - Store refrigerated
2. Mueller Hinton Agar (MHA)

Other Materials

Forceps
Swabs
Inoculating loop

Procedure

1. Pick one colony from CHOC, taking care not to carry over any agar from the medium.
2. Inoculate MHA and streak over the entire surface of the plate using a sterile swab.
3. Place X, V and XV disks on the surface of the agar in the form of a triangle with at least 30-35 mm between disks.
4. Incubate CO₂, 35°C X 18-24 hr.
5. Examine the pattern of growth around and/or between the disks.

Interpretation

Growth around the V and XV or the X and XV indicates a requirement for the single growth factor V or X respectively. Growth around only the XV disk indicates a requirement for both factors.

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Precautions

1. Avoid carry-over of growth factors.

Quality Control

Known positive and negative controls must be set up in parallel with the test.

H. influenzae (ATCC 35056): Growth around the XV disk only
H. parainfluenzae (ATCC 7901): Growth around V and XV disks

References

1. Difco Package insert, June 1984.