



X Factor discs

DD020

Used for the presumptive identification of *Haemophilus* species on the basis of their requirements for X or V factors or both.

Directions

Inoculate the surface of a Blood Agar (M073) plate or Brain Heart Infusion Agar (M211) plate with the test organisms by either streaking or surface spreading. Aseptically place the X (DD020), V (DD021) and X+V (DD022) factor discs on the plate, in the following positions:

Disc Position on the Agar plate

X factor disc 12 O' clock

V factor disc 4 O' clock

X+V factor disc 8 O' clock

Incubate the plates at 35 - 37°C for 24 - 48 hours. Observe for the growth in the neighbourhood of the discs.

Principle And Interpretation

Both X and V factors are growth factors that are essential for certain organisms like *Haemophilus* species and also enhance growth of organisms like *Neisseria* species.

X-factor discs are the sterile filter paper discs impregnated with growth factor X which are used for differentiating *Haemophilus* species in conjunction of V factor & X+V factor discs. *Bordetella* and *Haemophilus* species can also be identified on the basis of the requirement of X and V growth factors in the basal medium.

The X factor (hemin) and V factor (Coenzyme- Nicotinamide adenine dinucleotide NAD+) are impregnated on the sterile filter paper discs of diameter 6 mm.

The test organism requiring X factor alone, grows only in the vicinities of X and X+V factor discs. Those which require V factor alone grow in the vicinities of V and X+V factor discs. If both X and V factors are required, then the organism will grow only in the vicinity of the X+V factor discs. This satellite growth is seen around the disc promoting growth (1).

Quality Control

Appearance

Filter paper discs of 6 mm diameter bearing letters "X" in continuous printing style.

Cultural response

Cultural characteristics observed on Brain Heart Infusion Agar (M211) or Blood Agar Base (M073) after an incubation of 24-48 hours at 35-37°C.

Cultural Response

Organism	Growth with X factor	Growth without growth factor
Cultural response		
<i>Bordetella pertussis</i> ATCC 8467	Positive(initial isolation on Bordet Gengou Agar (M175))	Positive(initial isolation on Bordet Gengou Agar (M175))
<i>Haemophilus influenzae</i> ATCC 35056	Negative	Negative
<i>Haemophilus parainfluenzae</i> ATCC 7901	Negative	Negative

<i>Haemophilus</i> <i>haemoglobinophilus</i> ATCC19416	Positive	Negative
<i>Haemophilus ducreyi</i>	Positive	Negative

Storage and Shelf Life

Store between 2-8°C. For prolonged use store at -20°C. Use before the expiry date on the label.

Reference

1.Murray PR, Baron EJ, Jorgensen J.H., Pfaller M A, Tenover F.C., Tenover F.C.(Eds.),8th ed, 2003, Manual of Clinical Microbiology, ASM, Washington D.C.

Note:

Use known strains of *Haemophilus influenzae* to monitor the performance of the differentiation discs and the medium.

Do not use too heavy suspension of the test organisms as X or V factor carryover from the primary growth medium may take place

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