63237 TB-Medium Base according to Loewenstein-Jensen (Loewenstein-Jensen Medium, Base)

The selective Lowenstein Jensen Medium is used for isolation of Mycobacteria sp. from contaminated specimens. Egg supplement enhances Mycobacteria growth. Malachite green, penicillin and nalidixic acid inhibit bacteria Gram negative and Gram positive.

Composition:

Ingredients	Grams/**1.6** Litre
L-Asparagine	3.6
Monopotassium Phosphate	2.4
Magnesium Sulfate	0.24
Magnesium Citrate	0.6
Potato Flour	30.0
Malachite Green	0.4

Store prepared media below 8°C, protected from direct light. Store dehydrated powder, in a dry place, in tightly-sealed containers at 2-25°C.

Directions :
Suspend 37,2 g in 600 ml of purified water containing 12 ml of glycerol (49769). Heat with frequent agitation and boil for one minute. Sterilize at 121°C for 15 minutes. Cool to 45-50°C and, aseptically, add 1 L of homogenised sterile eggs and if required 4 vials of Gruft Mycobacterial Supplement (51803). Dispense into sterile culture tubes and coagulate at 85°C for 45 minutes in slant position.

Principle and Interpretation:
The original formulation of Löwenstein[1] was modified by Jensen[2] and Gruft[4,5] modified Loewenstein-Jensen Medium further with addition of two antimicrobial agents. The egg base medium supports a wide variety of Mycobacteria and can also be used for niacin testing.[6]
Malachite Green, Penicillin (Gruft Mycobacterial Supplement) and Nalidixic acid (Gruft Mycobacterial Supplement) prevents growth of the most contaminants surviving decontamination of the specimen while encouraging earliest possible growth of Mycobacteria. The RNA in the supplement acts as stimulant and help to increase the isolation rate of Mycobacteria. Malachite Green serves as an inhibitor and as pH indicator. Formation of blue zone indicates a decrease in pH by gram-positive contaminants (e.g. Streptococci) and yellow zones of dye destruction by gram-negative bacilli. Proteolytic contaminants cause localised or complete digestion of medium. The addition of Gruft Mycobacterial Supplement is also recommended for isolation and cultivation of Nocardia species from sputum, gastric washings and other clinical materials.[8] Hardy et al.[3] recommended each specimen to be inoculated and incubated in triplicate.

a.) to identify saprophytes – room temperature 25°C

b.) to identify presence or absence of pigmentation by photochromogens (35°C in light and dark)

Routinely cultivation is carried out aerobically at 35°C.

Cultural characteristics after 2-4 weeks at 35°C with 5-10% CO_2.

Organisms (ATCC)	Growth	with Gruft Supplement
Mycobacterium tuberculosis (25618)	+++ (granular, rough, warty, dry, friable colonies)	++
Mycobacterium kansasii (12478)	+++ (photochromogenic, smooth to rough)	++
Mycobacterium gordonae (14470)	+++ (smooth, yellow, orange colonies)	++
Mycobacterium avium (25291)	+++ (smooth nonpigmented colonies)	++
Mycobacterium smegmatis (14468)	+++ (wrinkled, creamy white colonies)	++ (wrinkled, creamy white colonies)

References:

1. E. Löwenstein, Zentralb. Bacteriol., Parasitenkd. Infektionskr. Abt. I Org., 120, 127 (1931)
2. K.A. Jensen, Zentralb. Bacteriol., Parasitenkd. Infektionskr. Abt. I Org., 125, 222 (1932)
3. A.V. Hardy, et al, Am. J. Publ. Hlth. 48(1), 754 (1958)
4. H. Gruft, J. Bacteriol., 90, 829 (1965)
5. H. Gruft, Health. Lab. Sci., 8(2), 79 (1971)
6. H. Boisvert, Ann. Inst. Pasteur, 99, 600 (1960)
7. A.L. Vestal, Procedures for the isolation and identification of Mycobacterium, DHEW Publication No. CDC 79-8230, CDC, Atlanta (1978)
8. Remel Technical Information, Ti No. 8500, Lenexa, Kann. Regional Media Laboratories, 6/11/80
9. W. Kleitmann, Resistance and susceptibility testing for *Mycobacterium tuberculosis*, Clinical Microbiology Newsletter, 17, 65-69 (1995)

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.