

Sellers Differential Agar

Differential medium used in studies of Gram negative non-fermenting bacilli

Formula in grams per liter:

Gelatin Peptone	20,00	Sodium Chloride	2,00
Dipotassium Phosphate	1,00	Magnesium Sulfate	1,50
Sodium Nitrate	1,00	Yeast Extract	1,00
L-Arginine	1,00	Sodium Nitrite	0,35
Bromothymol Blue	0,04	Phenol Red	0,008
Bacteriological Agar	13,50	D-mannitol	2,00

Final pH: 6,7 ± 0,2 at 25 °C

Preparation:

Suspend 43,4 grams of the medium in one liter of water. Mix well. Heat with frequent agitation and boil for one minute. Dispense into test tubes and sterilize at 121° C (15 lbs psi) for 10 minutes. Cool the tubes in a slanted position with a slant length of 7-7,5 cm and a butt depth of 3,5-cm.

Important: Immediately before inoculation, add 0,15 ml or 2 drops of 50% aqueous solution of dextrose, allowing it to run down the side of the tube opposite to the slant.

Uses:

Sellers Agar is inoculated by stabbing with a needle to the base of the tube and streaking the slant. Incubate at 35°C for 24 hours. It is a very useful medium to identify and differentiate *Pseudomonas aeruginosa*, *Herellea vaginicola*, *Mima polymorpha* and *Alcaligenes fecalis*. To aid in the identification of the non-fermenters, other media such as OF Basal Medium, Indol Nitrate Medium, etc. should be used. *Mima* and *Herellea* (*Acinetobacter calcoaceticus*) morphologically resemble *Neisseria* and frequently are erroneously reported as causes of gonococcal urethritis and meningococcal (resistant to penicillin) meningitis.

The differentiation is based on the detection of fluorescence, glucose oxidation, production of nitrogen gas and pH changes. Under UV light only the *pseudomonas* exhibit fluorescence, which is stimulated by magnesium and mannitol in the medium. At times it is necessary to hold the tubes 2 days for *Pseudomonas* to produce a typical alkaline (blue color) reaction in the medium. After incubation, check for oxidation of glucose by the appearance of a yellow band, which can disappear after 24 hours.

Typical Reactions				
MICROORGANISMS	<i>Pseudomonas</i>	<i>Mima</i> ¹	<i>Herellea</i> ²	<i>A. Fecalis</i> and <i>Vibrio</i>
Color of Slant	Green	Blue	Blue	Blue
Color of Butt	Blue or no change	No change	No change	Blue or no change
Color of Band	Blue at times	Absent	Yellow	Absent
Flourescence on Slant	Yellow green	No	No	No
Nitrogen gas	Yes	No	No	No

¹ *A. calcoaceticus* var. *Lwoffii*

² *A. calcoaceticus* var. *Anitratus*

Microbiological Tests:

Microorganisms	Growth	Slide	Base	Strip	Flourescence
<i>Acinetobacter calcoaceticus</i> ATCC 19606	Good	Blue	Green	Yellow	-
<i>Acinetobacter lwoffii</i> ATCC 9957	Good	Blue	Blue	-	-
<i>Alcaligenes faecalis</i> ATCC 8750	Good	Blue-green	Blue-green	-	-
<i>Pseudomonas aeruginosa</i> ATCC 27853	Good	Blue-green	Blue-green	Blue	+

