

**TECHNICAL SHEET**

<b>B1476</b>	<b>KLIGLER IRON AGAR</b>				
<b>Formula</b>					
<b>Ingredients :</b>		<b>gms/lit.</b>			
Beef extract		3.00			
Yeast extract		3.00			
Casein enzymic hydrolysate		20.00			
Sodium chloride		5.00			
Lactose		10.00			
Glucose anhydrous		1.00			
Ferrous ammonium sulphate, 6H <sub>2</sub> O		0.50			
Sodium thiosulphate pentahydrate		0.50			
Phenol red		0.025			
Agar		15.00			
Final pH (at 25°C) : 7.4 ± 0.2					
<b>Directions :</b>					
Suspend 57.70 grams of dehydrated powder in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Mix well and distribute into tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Allow the tubes to cool in slanted position to form slopes with about 1-inch butts. Best reactions are obtained on freshly prepared medium. Do not use screw capped tubes or bottles.					
<b>Principle :</b>					
Kligler Iron Agar, in addition to casein enzymic hydrolysate, beef and yeast extract, contains lactose and glucose (dextrose), which enables the differentiation of species of enteric bacilli. Phenol red is the pH indicator, which exhibits a colour change in response to acid produced during the fermentation of sugars. The combination of ferric ammonium sulphate and sodium thiosulphate enables the detection of hydrogen sulfide production, which is evidenced by a black color either throughout the butt, or in a ring formation near the top of the butt.					
<b>QC Tests – (I) Dehydrated Medium</b>					
Colour :		Light yellow to pink			
Appearance :		Homogeneous Free Flowing powder			
<b>(II) Rehydrated medium</b>					
pH (post autoclaving/heating) :		7.4 ± 0.2			
Colour (post autoclaving/heating) :		Reddish orange to red			
Clarity (post autoclaving/heating) :		Clear to slightly opalescent			
<b>(III) Q.C. Test Microbiological</b>					
Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.					
MICROORGANISM (ATCC )	GROWTH	SLANT	BUTT	GAS	H <sub>2</sub> S
Escherichia coli (25922)	Luxuriant	A	A	+	-
Enterobacter cloacae (13047)	Luxuriant	A	A	+	-
Proteus vulgaris (13315)	Luxuriant	K	A	-	+
Salmonella typhi (6539)	Luxuriant	K	A	-	+
Salmonella enteritidis (13076)	Luxuriant	K	A	+	+
Klebsiella pneumoniae (13883)	Luxuriant	A	A	+	-
Shigella flexneri (12022)	Luxuriant	K	A	-	-
Pseudomonas aeruginosa (27853)	Luxuriant	K	A	-	-
Yersinia enterocolitica (27729)	Luxuriant	K	A	V	-
Enterobacter aerogenes (13048)	Luxuriant	A	A	+	-
Salmonella Paratyphi A (9150)	Luxuriant	K	A	+	-
Citrobacter freundii (8090)	Luxuriant	A	A	+	+
Key : A = acid production (yellow) K = alkaline reaction (red) + = positive or blacking V = variable					

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<b>Precautions :</b>	<p>1. For Laboratory Use.</p> <p>2. Follow proper, established laboratory procedures in handling and disposing of infectious materials.</p>				
<b>Limitations :</b>	<p>1. Since the nutritional requirements of organisms vary, some strains may be encountered that fail to grow or grow poorly on this medium.</p> <p>2. H<sub>2</sub>S – producing organisms may produce a black precipitate to such a degree that the reaction in the butt is completely masked. If H<sub>2</sub>S is produced, dextrose is fermented even if it is not observed.</p> <p>3. Further biochemical tests and serological typing must be performed for definite identification and confirmation of organisms.</p> <p>4. Do not use as inoculating loop to inoculate a tube of Kligler Iron Agar. While stabbing the butt, mechanical splitting of the medium occurs, causing a false positive result for gas production.</p> <p>5. Best reactions are obtained on freshly prepared medium.</p> <p>6. A pure culture is essential when inoculating Kligler Iron Agar. If inoculated with a mixed culture, irregular observations may occur.</p> <p>7. Hydrogen sulfide determinations using Kligler Iron Agar should be limited to the members of the Enterobacteriaceae. Other organisms may require more sensitive methods for detection of H<sub>2</sub>S production.</p> <p>8. Tubes should be incubated with caps loosened to allow a free exchange of air, which is necessary to enhance the alkaline condition on the slant.</p>				
<b>Use:</b>	It is recommended for identification of Pseudomonas species. It can also be used for the differential identification of gram-negative enteric bacilli on the basis of the fermentation of glucose, lactose and H <sub>2</sub> S production. Recommended by International Organization for Standardization (ISO), 1995, Draft ISO/DIS 13720.				
<b>Storage :</b>	Dehydrated medium- below 30°C Prepared medium– Between 2 to 8°C.				
<b>Packing :</b>	500 gm. bottle				
<b>Product profile:</b>	Reconstitution	Quantity on Preparation (500g)	pH (25°C)	Supplement	Sterilization
<b>B1476</b>	55.70 g/l	8.976 L	7.4 ± 0.2	NIL	121°C / 15 minutes