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X.L.D. MEDIUM CM0469		

## X.L.D. MEDIUM

CM0469

### Typical Formula\*

Yeast extract	grams per litre	3.0
L-Lysine HCl		5.0
Xylose		3.75
Lactose		7.5
Sucrose		7.5
Sodium desoxycholate		1.0
Sodium chloride		5.0
Sodium thiosulphate		6.8
Ammonium iron (III) citrate		0.8
Phenol red		0.08
Agar		12.5

\* adjusted as required to meet performance standards

### Directions

Suspend 53g in 1 litre of distilled water. With frequent agitation, bring to the boil to dissolve completely. Cool to 50°C. Mix well and pour into sterile Petri dishes. DO NOT AUTOCLAVE. DO NOT OVERHEAT.

### Physical Characteristics

Straw/pink, free-flowing powder  
 Colour on reconstitution - red  
 Moisture level - less than or equal to 7%  
 pH 7.4 ± 0.2 at 25°C  
 Clarity - clear  
 Gel strength - firm, comparable to 12.5g/litre of agar

### Microbiological Tests Using Optimum Inoculum Dilution

Control Medium: Tryptone Soya Agar

### Reactions after incubation at 37 ± 2°C for 24 ± 3 hours

Inoculation with mixed cultures using diminishing sweep technique

Medium is challenged with 1E+03 to 1E+05 colony-forming units (cfu) of *Salmonella* and *Shigella* spp. and 1E+05 to 1E+07 cfu for *Escherichia coli* ATCC® 8739.

*Salmonella abony*

NCTC6017

1-3mm red colonies, black centre

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<i>Salmonella enteritidis</i>	ATCC® 13076	1-2mm red colonies, black centre
<i>Salmonella typhimurium</i>	ATCC® 14028	1-2mm red colonies, black centre
<i>Salmonella virchow</i>	NCTC5742	1-2mm red colonies, black centre
<i>Salmonella arizonae</i>	ATCC® 13314	1-3mm red colonies, black centre
<i>Salmonella nottingham</i>	NCTC7832	1-3mm red colonies, black centre
<i>Shigella sonnei</i>	ATCC® 9290	0.5-7mm irregular/smooth red colonies
<i>Shigella flexneri</i>	ATCC® 12022	0.5-2mm irregular, red colonies

In mixed culture, using the diminishing sweep technique, a satisfactory result is represented by diagnostic reactions of Salmonellae and Shigellae strains and *Escherichia coli*. Clear differentiation must be seen and is based on the colour and morphology of the colonies.

Inoculation with pure cultures

Medium is challenged with 10-100 colony-forming units

<i>Pseudomonas aeruginosa</i>	ATCC® 9027	No growth or 0.5-2mm red colonies
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For *Pseudomonas aeruginosa* ATCC® 9027, a satisfactory result is represented by recovery equal to or less than 90% of the control medium.

<i>Proteus mirabilis</i>	ATCC® 12453	0.5-2mm orange/red colonies, with or without black centre, no swarming
<i>Proteus mirabilis</i>	ATCC® 29906	0.5-2mm orange/red colonies, with or without black centre, no swarming
<i>Serratia marcescens</i>	ATCC® 8100	1-2mm orange/yellow colonies
<i>Citrobacter freundii</i>	ATCC® 8090	0.5-2mm yellow colonies
<i>Klebsiella pneumoniae</i>	ATCC® 29665	2-4mm yellow, mucoid colonies

Other pure cultures are inhibited or shall produce colonies with a negative diagnostic reaction.

Medium is challenged with 1E+04 to 1E+06 colony-forming units

<i>Staphylococcus aureus</i>	ATCC® 6538	No growth
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Negative strains are inhibited.

Inoculation using diminishing sweep technique

Medium is challenged with 1E+04 to 1E+06 colony-forming units

<i>Escherichia coli</i>	ATCC® 11775	No growth or 0.5-4mm yellow colonies
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*Escherichia coli* ATCC® 11775 is inhibited or shall produce colonies with a negative diagnostic reaction.

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Medium is challenged with 1E+04 to 1E+06 colony-forming units

*Shigella sonnei*                      ATCC®25931                      0.5-7mm irregular/smooth red colonies

*Shigella sonnei* ATCC®25931 shall produce colonies with a positive diagnostic reaction.

Equivalent results are obtained after incubation at 30-35°C for 24 hours.

#### Testing performed in accordance with ISO11133:2014

#### Reactions after incubation at 37 ± 2°C for 24 ± 3 hours

Medium is challenged with 50-120 colony-forming units

<i>Salmonella enteritidis</i>	ATCC®13076	WDCM00030	1-3mm red colonies, black centre
<i>Salmonella typhimurium</i>	ATCC®14028	WDCM00031	1-3mm red colonies, black centre

A satisfactory result is represented by recovery of equal to or greater than 70% of the control medium.

Medium is challenged with 1E+04 to 1E+05 colony-forming units

<i>Escherichia coli</i>	ATCC®8739	WDCM00012	No growth or 0.5-4mm yellow cols
<i>Escherichia coli</i>	ATCC®25922	WDCM00013	No growth or 0.5-4mm yellow cols


Inhibited strains shall produce no growth or at least a 1 log (10) reduction with a negative diagnostic reaction when compared to the control medium.

Inoculation using diminishing sweep technique

Medium is challenged with 1E+04 to 1E+06 colony-forming units

<i>Enterococcus faecalis</i>	ATCC®29212	WDCM00087	No growth
<i>Enterococcus faecalis</i>	ATCC®19433	WDCM00009	No growth

Negative strains are inhibited.

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Testing performed in accordance with current CLSI M22 A

#### Reactions after incubation at 35°C for 18-24 hours

Medium is challenged with 10-100 colony-forming units

<i>Shigella flexneri</i>	ATCC®12022	0.5-2mm irregular, red colonies
<i>Salmonella typhimurium</i>	ATCC®14028	1-2mm red colonies, black centre

A satisfactory result is represented by recovery of equal to or greater than 70% of the control medium.

Medium is challenged with 1E+04 to 1E+06 colony-forming units


<i>Enterococcus faecalis</i>	ATCC®29212	No growth
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Negative strains are inhibited.

Medium is challenged with 1E+04 to 1E+06 colony-forming units

<i>Escherichia coli</i>	ATCC®25922	No growth or 0.5-4mm yellow cols
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Inhibited strains shall produce no growth or at least a 1 log (10) reduction with a negative diagnostic reaction when compared to the control medium.

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## Revision History

Section / Step	Description of Change	Reason for Change	Reference
Microbiological Tests	Update to specification for <i>Shigella sonnei</i>	Change control	BT-CC-1911
Microbiological Tests	Salmonella and Shigella mixed culture testing changed from low number quantitative to high number qualitative testing.	Change control	BT-CC-2398