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# WILLIS -HOBBS AGAR

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Works for me

[dx.doi.org/10.17504/protocols.io.3exgjfn](https://dx.doi.org/10.17504/protocols.io.3exgjfn)

SoWa RI Anaerobic and Molecular Microbiology (public)

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## ABSTRACT

For the diferential isolation of Clostridium sp.

## EXTERNAL LINK

<http://himedialabs.com/TD/M1375.pdf>

## DOI

[dx.doi.org/10.17504/protocols.io.3exgjfn](https://dx.doi.org/10.17504/protocols.io.3exgjfn)

## PROTOCOL CITATION

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MANUSCRIPT CITATION please remember to cite the following publication along with this protocol

## EXTERNAL LINK

<http://himedialabs.com/TD/M1375.pdf>

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## CREATED

May 28, 2019

## LAST MODIFIED

Sep 09, 2020

## PROTOCOL INTEGER ID

23735

## GUIDELINES

taken from: <http://himedialabs.com/TD/M1375.pdf>

#### MATERIALS

NAME	CATALOG #	VENDOR
Agar		
Peptone A FROM MEAT	G213.SIZE.500g	Bio Basic Inc.
D-Lactose, monohydrate	LC4280.SIZE.500g	Bio Basic Inc.
Neutral red	NB0652.SIZE.25g	Bio Basic Inc.
Sodium chloride	S3014	Sigma Aldrich
Meat Extract	70164-100G	Merck Millipore
Egg Yolk Emulsion	414722,1608	

#### SAFETY WARNINGS

Use temperature resistant gloves for taking the media out of the autoclave and during pouring of the petri dishes.

#### BEFORE STARTING

- Make a 10% skim milk solution and sterilize it by autoclaving at 15 lbs pressure **121 °C** for **00:15:00**.
- Turn ON the water bath to **50 °C**
- Before taking the media from the autoclave, open the petri dish bags and organize them on the flow box. Mark the petri dishes as W-H agar.

- For the preparation of 1L of media (50 to 66 petri dishes depending of depth) weigh:

**10 g Peptic Digest from Animal tissue**

**10 g Meat Extract**

**5 g NaCl**

**12 g Lactose**









**0.032 g Neutral Red**

**10 g Agar**

**30 mL Egg yolk Emulsion**

**Final pH ( at 25°C) 7.0±0.2**

- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 lbs pressure **121 °C** for **00:15:00**.

- 4 Cool to  **50 °C**
- 5 Add  **120 mL** sterile skimmed milk 10% solution.
- 6 Aseptically add  **30 mL** Egg Yolk Emulsion.
- 7 Pour  **15 mL** or  **20 mL** of media in petri dishes depending on desired depth .
- 8 Let cool the open petri dishes under UV light for  **00:10:00** to  **00:15:00** .
- 9 Close petri dishes and keep at  **4 °C** until use. (2 weeks max)