

Aug 14, 2024

Acidified 0.5x Potato Dextrose Agar (APDA)

DOI

dx.doi.org/10.17504/protocols.io.j8nlk82jdl5r/v1

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DOI: dx.doi.org/10.17504/protocols.io.j8nlk82jdl5r/v1

Protocol Citation: Gabriela Paredes 2024. Acidified 0.5x Potato Dextrose Agar (APDA). **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.j8nlk82jdl5r/v1>

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Protocol status: Working

We use this protocol and it's working

Created: August 14, 2024

Last Modified: August 14, 2024

Protocol Integer ID: 105297

Keywords: APDA, Plant Pathology, microbiology, cultivation of fungi

Funders Acknowledgement:

USDA

Grant ID: 2021-67013-33943



Disclaimer

This protocol is provided for research and educational purposes only. Ensure that all procedures are conducted by trained personnel following relevant safety and ethical guidelines. The authors and publishers are not responsible for any injuries, damages, or legal consequences resulting from the use of this protocol.

Abstract

Acidified 0.5x Potato Dextrose Agar (APDA) is a specialized culture medium used in microbiology for the selective cultivation of fungi, particularly in environments where lower pH is required to inhibit bacterial growth. This medium contains a reduced concentration of nutrients compared to standard PDA, making it useful for specific fungal isolations and studies.

APDA is not ideal for cultivating bacteria, fungi that prefer neutral pH environments, or nutrient-demanding fungi. It should also be avoided in situations where accurate quantification of fungal growth, enzyme activity studies, or broad-spectrum fungal isolation is required.

Guidelines

- **Aseptic Technique:** Ensure strict aseptic conditions throughout the preparation and handling of the medium to prevent contamination. This includes working under a laminar flow hood when pouring the medium into Petri dishes.
- **Chemical Safety:** Handle all chemicals, especially 85% lactic acid, with appropriate personal protective equipment (PPE) including gloves, lab coats, and eye protection. Be aware of the potential hazards associated with concentrated acids.
- **Sterilization:** Verify that the autoclave is functioning correctly and that all materials are properly sterilized to avoid cross-contamination. Allow the autoclave to cool completely before opening to avoid steam burns.
- **Homogeneous Mixing:** Ensure that all components are thoroughly dissolved and mixed before and after autoclaving, as well as after the addition of lactic acid, to achieve a uniform medium.
- **Temperature Control:** Monitor the temperature closely after autoclaving. Pour the medium into Petri dishes once it has cooled to between 55°C and 40°C to avoid issues with condensation and to ensure proper gelation.
- **Disposal:** Dispose of all waste materials, including unused medium and contaminated plates, according to institutional biohazard waste disposal protocols. Autoclave all biological waste before disposal to prevent the release of potentially harmful organisms.

Materials

- 10 g of **potato dextrose broth**
- 16-18 g of **agar**
- 1 ml of **85% lactic acid**
- Distilled water (1 liter)
- Sterile Petri dishes
- Stir bar

Equipment

Adventurer™ Analytical Balances	NAME
Analytical balance	TYPE
Ohaus	BRAND
30100600	SKU
https://www.fishersci.com/shop/products/ohaus-adventurer-analytical-balances-7/p-4918285 ^{LINK}	



Equipment

8-Liter Autoclave

NAME

Portable Stainless Steel Pressure Steam Sterilizer

TYPE

China

BRAND

XFS-D-8L

SKU

<https://www.dentalplaza.co.uk/Dentist-8L-Portable-Steam-Autoclave-Sterilizer-168696-dental.html>^{LINK}

Voltage: 220 V (AC)

SPECIFICATIONS

Power: 1.2 kW

Working Medium: Steam

Design Pressure: 0.17 MPa

Working Temperature: 129 °C

Frequency: 50 Hz

Useful Life: 5 Years

Delivery Date: 3. Oct, 2019



Equipment

Magnetic Stirrer RT touch series

NAME

Magnetic Stirrer

TYPE

Thermo Fisher

BRAND

88880013

SKU



Safety warnings

1. **Handling Lactic Acid:** 85% lactic acid is highly concentrated and corrosive. Wear appropriate personal protective equipment (PPE) such as gloves, lab coats, and eye protection when handling to avoid skin contact or inhalation.
2. **Sterilization Safety:** Ensure that the autoclave is properly secured before running the cycle. After autoclaving, handle the hot flask with heat-resistant gloves to avoid burns.
3. **Aseptic Technique:** Maintain aseptic conditions during all steps, particularly after the autoclaving process, to prevent contamination.

Ethics statement

This protocol for preparing and using Acidified 0.5x Potato Dextrose Agar (APDA) is intended for use in research and educational settings. Researchers are responsible for ensuring that all work conducted with this medium adheres to institutional and national biosafety guidelines and ethical standards. This includes the proper handling, disposal, and sterilization of microbial cultures to prevent contamination and environmental release. Any work involving potentially pathogenic fungi should be conducted in a biosafety level-appropriate facility, with all necessary precautions taken to protect laboratory personnel and the environment. The use of this protocol must comply with all applicable laws and regulations governing the study and manipulation of microorganisms.



Before start

Ensure that all materials and equipment are available, clean, and sterilized. Prepare the lactic acid solution and ensure that the autoclave is set up and ready for use.




Prepare the Medium:

- 1 In a 1-liter Erlenmeyer flask with a stir bar, dissolve the following components in 1 liter of distilled water (dH₂O):

-  10 g of potato dextrose broth.
-  18 g of agar.

Stir the mixture thoroughly until all components are fully dissolved.

Sterilize:




- 2 Autoclave the solution at  121 °C for 20minutes using the liquid cycle to sterilize the medium.

20m

- 3 After autoclaving, remove the flask from the autoclave and place it on a magnetic stirrer.

Stir the medium for approximately 1 minute until the solution is homogeneous.

Acidification:

- 4
 - Add  1 mL of **85% lactic acid** to the medium.
 - Stir the medium again for approximately 1 minute to ensure the lactic acid is evenly distributed.
 - Place the flask in a room temperature water bath and stir continuously until the temperature of the medium cools to between  55 °C and  40 °C .

Pour into Petri Dishes:

- 5
 - Work aseptically to pour the cooled medium into sterile Petri dishes.
 - Allow the medium to solidify at room temperature before storing or using.