



FOOD SAFETY LAB / MILK QUALITY
IMPROVEMENT PROGRAM

Standard Operating Procedure



Title: Remelting Agar Media

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Author: Sherry Roof

Approved by: Martin Wiedmann

Remelting Agar Media

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SECTION 1 INTRODUCTION

1.1 Purpose

The purpose of this document is to set forth **standard** guidelines for re-melting solid agar media.

1.2 Scope

This SOP applies to the Food Safety Lab, including the Laboratory for Food Microbiology and Pathogenesis of Foodborne Diseases and the MQIP Lab.

1.3 Definitions

1.4 Safety

Never tightly seal containers. This results in pressure build-up and is an explosion hazard.

The autoclave heats to 121°C. Wear appropriate Personal Protective Equipment (PPE) including a lab coat, heat resistant gloves, and eye protection when loading and unloading the autoclave.

The boiling water bath contains water and steam at 100°C. Wear appropriate Personal Protective Equipment (PPE) including a lab coat, heat resistant gloves, and eye protection when placing media into the water bath and removing it once melted.

When preparing media that will be allowed to solidify to be re-heated later, use only approved laboratory borosilicate tempered glassware (e.g., Pyrex, Kimax, or equivalent) without any cracks, chips, or scratches. Over time and with damage, glassware can lose temper, which may result in an explosion.



SECTION 2 MATERIALS

- Corningware bottles and caps
- Agar media
- Autoclave
- PPE
- Water bath
- Boiling water bath – Grant SBB Aqua 26 Plus, located at the end of bench 23 between rooms 356 and 358 of the FSL/MQIP lab



SECTION 3 PROCEDURES

Melting Agar Media in the Autoclave

1. Place the Corning bottle in a secondary, autoclavable vessel.
2. Loosen the cap until it is only gripping the first thread of the bottle.
3. Place the vessel in the autoclave.
4. Select the 5-minute heat cycle on the programming pad, then “Run” (Note: The entire cycle will take +/- 15 minutes, so plan accordingly).
5. Check the water volume in the water bath; add water if needed. Turn the bath on, set the temperature to 55°C. Check back prior to placing media in bath to ensure temperature is correct.
6. When the autoclave signals the end of run, check the information display. DO NOT try to open the door until the readout says “Open Door”.
7. Using hot gloves, carefully remove your media and take it to the water bath.
8. Remove the water bath cover, standing clear of the released steam. Check that the temperature is correct. Carefully place the bottle of media into the 55°C water bath. Leave media in this water bath for at least 15 minutes to allow it to cool to a safe temperature for use.
9. Remove the bottle of media from the 55°C water bath and allow the bottle to sit on paper towels to drain, then wipe with 70% ethanol before use. Turn off the water bath.

3.2. Melting Agar Media in the Boiling Water Bath

1. Check the water volume in the boiling water bath; add water if needed. Water level should exceed the level of the media in the bottle by at least 4 mm. Turn the bath on and set the temperature to the maximum setting.
2. Check the water volume in a second water bath; add water if needed. Turn the bath on and set the temperature to 55°C. Check back prior to placing media in bath to ensure temperature is correct.
3. Loosen the media bottle cap(s) until it is only gripping the first thread of the bottle(s).
4. Place the media bottle(s) into the boiling water bath. If the bottle floats, it can be sunk using a lead weight.
5. Once the water in the boiling water bath has heated to boiling, carefully check the media to see if it has completely melted approximately every 5 minutes.
6. Using hot gloves, carefully remove your media from the boiling water bath once fully melted and place it in the 55°C water bath. Turn off the boiling water bath. Leave media in the 55°C water bath for at least 15 minutes to allow it to cool to a safe temperature for use.
7. Remove the bottle of media from the 55°C water bath and allow the bottle to sit on paper towels to drain, then wipe with 70% ethanol before use. Turn off the 55°C water bath.



SECTION 4 TROUBLESHOOTING

To protect the integrity of the media, it should only be re-melted once.

There have been instances when contamination present in the water bath is transferred to samples placed in the bath. To guard against this, allow the bottle to sit on paper towels to drain, then wipe with a 70% ethanol cloth.

SECTION 5 REFERENCES

SECTION 6 METHOD VERSION & CHANGES

VERSION	DATE	EDITOR	COMMENTS
Version 1	12/19/2019	ser15	Original SOP
Version 2			
Version 3			