

SOP-M006

Bacteria in Ground Beef using the Time Lapse Robot / Bardot

Objective: To view various stages of bacteria growth using raw hamburger

For Standard USDA size sample 25g of ground beef

Buy refrigerated ground beef from commercial source, 85% lean variety. Write down dates from package.

Allow TSB broth to come to room temperature. For a full sized sample you will need 225 ml of TSB.

Keep ground beef in the refrigerator until you are ready to start.

Weigh out 25g of ground beef.

Place the weighed out beef into a stomaching bag.

Add 225 ml of TSB to stomaching bag.

Stomach at medium speed for 1 minute at room temperature. Pour the sample into a 50cc tube. Let it settle for a few minutes. Then collect 100ul from the 50cc tube and place it in the 4mls of sterile PBS tube (1). From that tube remove 100ul and place it in the second tube(2) containing 4mls of PBS. Finally, remove 100ul from tube(2) and placing it in an eppendorf tube containing 900ul of PBS.

After dilution procedure, plate immediately onto TSA plates (50ul from the eppendorf tube). Expected CFU/g of ground beef is typically high 10^4 , range varies from 10^3 to 10^5 .

Incubate at 30 C. **Note: not 37 C because much of the associated flora doesn't grow above 30C.**

For 1/10 scale sample (ratio of ground beef to TSB is 1:9)

Buy refrigerated ground beef from commercial source, 85% lean variety. Write down dates from package.

Allow TSB broth to come to room temperature. For a sample you will need 22.5 ml of TSB.

Keep ground beef in the refrigerator until you are ready to start.

Weigh out 2.5g of ground beef.

Place the weighed ground beef into a stomaching bag.

Add 22.5 ml of TSB to stomaching bag.

Stomach at medium speed for 1 minute at room temperature. Pour the sample into a 50cc tube. Let it settle for a few minutes. Then collect 100ul from the 50cc tube and place it in the 4mls of sterile PBS tube (1). From that tube remove 100ul and place it in the second tube(2) containing 4mls of PBS. Finally, remove 100ul from tube(2) and placing it in an eppendorf tube containing 900ul of PBS.

After dilution procedure, plate immediately onto TSA plates (50ul from the eppendorf tube). Expected CFU/g of ground beef is typically high 10^4 , range varies from 10^3 to 10^5 .

Incubate at 30 C. **Note: not 37 C because much of the associated flora doesn't grow above 30C.**

Created by: George (USDA)

Date:

Verified by: _____ Date: _____

Print Name

Sign Name