Honey and Biotech

Is raw honey sterile?

General: Honey is the oldest wound dressing material known to humans. Per the National Institute of Health, studies reported that honey has antioxidant, antibacterial and anti-inflammatory properties. It can be used as a wound dressing to promote rapid and improved healing. These effects are due to honey's antibacterial action. Healing is also promoted due to its high acidity, osmotic effect, antioxidant content and hydrogen peroxide content. This experiment initiates the investigation of raw honey and its sterility by using serial dilution and spread plating methodology to enumerate the number of bacteria within a raw sample.

Vocabulary

Define the following words in your own terms and cite sources used:

serial dilution

CFU

antibacterial

antioxidant

anti-inflammatory

raw honey

Problem: Is raw honey purchased from local markets sterile?

Hypothesis:

Variables:

Independent:

Dependent:

Controls:

Safety: eye protection

Materials

nutrient agar plates as prepared by SLOP#99 (in Soy Fresh Unit found at grownextgen.org) or already prepared plates
distilled water
micro tubes
H1000 micropipette
H200 micropipette
micropipette tips
honey samples

Tools & equipment: vortexer, electronic balance, incubator

PROCEDURE

Prep Honey Sample:

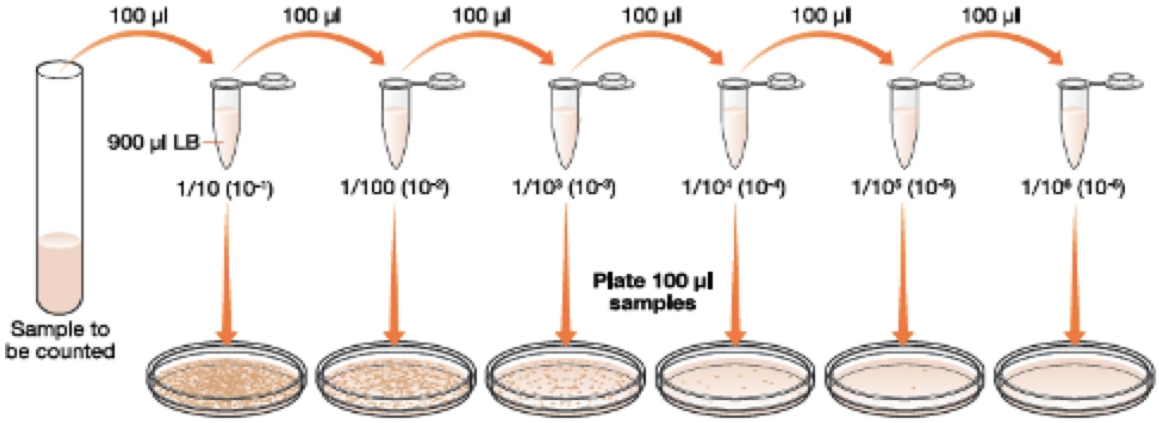
1. Label a micro tube as 10^0; add initials and honey sample number.
2. Using a H1000 micropipette, add 900µL of distilled water into the labeled micro tube.
3. Using a H200 micropipette, add 100µL of honey sample to the micro tube with distilled water and mix for 15 seconds on a vortexer. (If honey is too thick, warm in a 60°C water bath for 2-3 minutes before pipetting.)
4. Repeat steps 1-3 for each honey sample being investigated.

Set Up Serial Dilutions

1. Label micro tubes 10^1 through 10^3; add initials and honey sample number.
2. Use a H1000 micropipette, to add 900µL of distilled water into each micro tube.
3. Then use a H200 micropipette to add 100µL from the 10^0 tube to tube labeled 10^1 and mix on the vortexer for 15 seconds.
4. Using aseptic technique, transfer 100µL of sample in 10^1 tube to the labeled 10^2 tube and mix on the vortexer for 15 seconds.
5. Using aseptic technique, transfer 100µL of sample in 10^2 tube to the labeled 10^3 and mix on the vortexer for 15 seconds.

Set Up Spread Plates

1. Along the outer rim of the bottom of each nutrient agar plate, label the plate with corresponding dilution factor, honey number, date and initials.
2. Using aseptic technique, pipet 100µL of each diluted sample in the middle of the corresponding plate.
3. Using a plate spreader or sterile swab, move the sample over the entire plate. If using a sterile swab, use a new one on each plate. If using a plate spreader, be sure to rinse the plate spreader in between each sample.
4. Example diagram for setting up serial dilutions and plating: (adapted from Bio-Rad)



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Data

Create a data table in APA Format

Conclusion

REE: (Restate evidence; discuss if hypothesis is accepted, rejected or inconclusive and give actual data evidence to support)

PE: (Possible/Potential errors that occurred during the investigation -- reminder your lab partner is not an error)

PA: (Practical applications; importance of investigation)

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