Effectiveness of biobased cleaners

Standard Operating Procedure

Laboratory: Biotech/Bioresearch/Food Science
Location: Food Science Lab
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General: Microbes play an integral role in lab safety and quality. The purpose of this protocol is to investigate the effectiveness of various types of honey varieties that may have antimicrobial action.

Safety: Safety Glasses

Materials

E. coli K12 starter plates (microbial culture)
single hole punch
alcohol burner or Bunsen burner
sterile petri dishes
incubator
sterile swabs
honey varieties
filter paper
70% IPA
inoculating loop
sterile forceps
nutrient agar
sharpie
rulers or calipers

Procedure

1. Prepare nutrient agar plates per Nutrient Agar Preparation SOP
2. Sterilize single-hole punch by dipping in 70% alcohol, then running through the fame of alcohol burner or Bunsen burner.
3. Label petri dishes with names of honey varieties to be tested.
4. With sterile hole-punch, punch out 4-5 discs from filter paper onto labeled sterile petri dishes.
5. Add 1 ml of honey over punched discs in appropriately labeled petri dishes and allow discs to soak 1 minute in honey.
6. Using sterilized forceps, remove discs from honey and place into newly labeled sterile petri dishes to dry. Make sure petri dishes containing the soaked discs are placed next to a lit alcohol or Bunsen burner when drying to create an updraft limiting aerial contamination.
7. While discs are drying, label the bottoms of 5 petri dishes with nutrient agar by dividing the base of the plate into 4 quadrants drawing a cross with a marker. Label quadrants 1-4 along the edge of the plate.
8. Next, label along the edge of the base of petri dishes with the type of honey, technicians' initials and date.
9. Flame an inoculating loop using an alcohol or Bunsen burner to sterilize. Select a colony from the *E. coli K12* starter plate to swab first test plate. Repeat this step until each test plate has been swabbed with *E. coli K12*.
10. Using sterile forceps, place the dry treated paper disc in the middle of each quadrant of the appropriately labeled plates. Flame forceps each time to prevent

cross-contamination.

11. Seal the edge of each petri dish with parafilm and place each petri dish upside down on shelf of incubator.
12. Incubate plates for 24 hours at 37°C.
13. After 24 hours of incubation, remove plates from incubator and measure the ring of inhibition of each trial.
14. Record.