Unknown bacteria

Science, Biology

In order to treat a disease correctly and efficiently, it is important to first identify the microbe that is causing the disease. Isolating the bacteria that is causing the disease and using an antibiotic that specifically targets that microbe, can help prevent or reduce the overuse of broad-spectrum antibiotics-which can lead to antibiotic resistance. When trying to determine the type of microbe that is causing a disease, there are a number of biochemical tests that are administered.

The results of these ifferent tests are analyzed against the characteristics of different bacteria and the perpetrator is revealed. Method I subjected my unknown bacteria to 7 different biochemical tests in order to identify it. I first performed a streak plate in order to get an isolated colony that I could inoculate and perform the tests on. The Sulfur, Indole and Motility test (SIM), the Glucose Fermentation test and the Urea test to help identify my unknown bacteria.

For the SIM test, I aseptically inoculated the bacteria into a tube of the SIM media and let it incubate until the next class period. I took the tube out of the incubator and irst observed it for the presence of sulfur as well as for motility. I then added 4 drops of Kovac's reagent to the SIM agar deep and observed the reaction. In this test, there are more than one enzyme and substrate involved. The sulfur part of this test the enzymes involved are Cysteine Desulfurase and thiosulfate reductase. Cysteine and thiosulfate are the available substrates that are in the media.

If either enzyme is produced by the bacteria being inoculated, Hydrogen Sulfide will be produced and will combine with the Iron in the Ferrous sulfate

that is already in the media and will roduce a black precipitate-which is a positive result. No production ofa black precipitate is a negative result. In the Indole part of this test, the enzyme involved is tryptophanase has been produced, I added Kovac's reagent to the media and observed it for a red color-which would indicate a positive result. No appearance of red would be a negative result.

Another test I used to identify my unknown was the Glucose Fermentation test. This test determines whether the bacteria produces an acid, acid and gas or an alcohol. For this test, I inoculated my bacteria into a tube of Glucose broth that contained a Durham tube. If, after incubation, there is a bubble in the Durham tube, that is a positive result for the production of gas. No bubble would be a negative result. The media in this tube contained the indicator Phenol Red. A positive result for fermentation to an acid is a yellow color. A negative result for acid production is red.

The Urea test was another test I performed to try and identify my bacteria. For this test I inoculated my unknown into a tube of urea broth. I let them incubate until the next class period. Urease is the enzyme involved in this test that breaks down urea. Phenol red is the indicator used in this test in order to etect a change in PH. If the pH is less than 7, the Phenol Red is yellow-which is a negative result. If the pH is greater than 7, that indicates the presence of the enzyme urease and the Phenol Red will be hot pink-which is a positive result. Forrest & Elliott, 2012) Results Test/CultureEscherichia coli Proteus vulgaris Klebsiella pneumonia Shigella flexneri Salmonella typhimurium Unknown #9 Sulfur Indole +1- Motility

Methyl Red Voges-Proskauer Citrate Glucose Lactose Urea Phenylalanine Deaminase My unknown bacteria did not give off a black precipitate in the Sulfur portion of the SIM test so that was a negative result. It turned a pink color in the Indole portion of the SIM test so that was a positive result. The SIM media was cloudy around the area where I inoculated the bacteria, indicating it is positive for motility.

The Methyl Red test produced an orange color, which was a negative result. The VP part of the MR-VP test produced an orange color as well, which is a negative result. There was no growth on the Simmon's Citrate media, indicating a negative result for that test. On both the Glucose and Lactose test, the broth was yellow and there was a bubble, which indicated that the bacteria produced and acid and a gas.