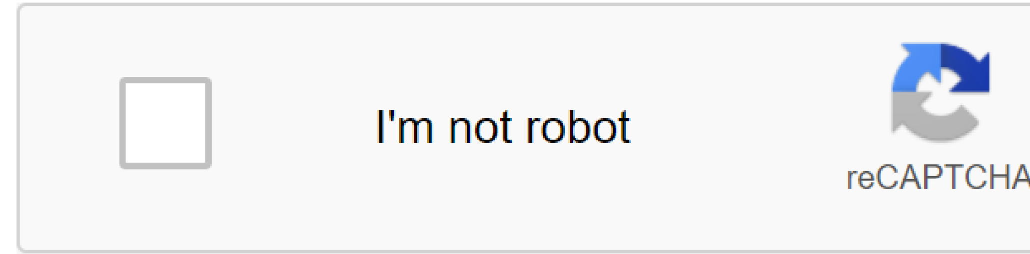
Pure culture methods pdf



Continue

The term Pure Culture refers to the population or colony of cells that grow in the absence of other species or types. The idea is that if agar plates are grafted with only one species and there is no contamination, then each colony on the plate will consist of genetically identical cells that came from a single progenitor or parent cell. The technique of pure culture allows isolating one species from mixed culture is a useful tool that helps to get a single body look from a mixed culture. There are two common methods of pure culture. Both of these methods lead to separate colonies that are isolated from mixed culture. Pour the stove - pour the plates strip plate. Color colony Gram spot results. S. aureus bacterial colonies Brooke Bearden - Own work, CC BY-SA 4.0, Bill Branson - (edited by Fir0002) (Edited by Drhx) - This image was released by the National Cancer Institute, the agency part of the National Institutes of Health, with ID 2230 (image) (next). Bill Branson - (Edited by Fir0002) (Edited by Drhx) - This image was released by the National Cancer Institute, an agency comprised of the National Institutes of Health, with ID 2230 (image) (next). Public domain, 1 2 3 Mixed Culture in Broth You only have one shot at getting this right because the TSA Agar will harden quickly. There are all fully prepared and ready to go before you get TSA pipes. Get 3 empty plates (petri dishes). Label them 1, 2 and 3 along with, names and lab days and time and body. Get a broth containing mixed bacterial culture and a loop and flame loops. Get one molten TSA tube (TSA Tube 1) from the water bath and immediately transfer one loop of mixed culture broth to the molten TSA tube (TSA Tube No. 1) Use a loop to mix for 5 seconds. Have your partner get another molten TSA tube (TSA Tube 2) from the water bath and immediately transfer one loop mixture from the first molten TSA tube (TSA Tube 1) to the second molten TSA tube (TSA Tube 2). Use a loop to mix for 5 seconds. Immediately pour the contents of the TSA 1 tube into the Petri dish labeled 1. Have your partner get a third molten TSA tube from the water bath (TSA Tube 3) and immediately transfer one loop mixture from the second molten TSA tube (TSA Tube 2) to the third molten TSA tube (TSA Tube 3). Immediately pour the contents of the TSA 2 tube into a petri dish labeled 2.Immediately pour the contents of the tsa tube 3 into the petri dish labeled 3. Gently rotate the plate to help the media spread until hardened. Once your plates harden, incubate them with your feet on to avoid condensation. You'll need your strip of plates and pour the plate and three three inclinations to isolate your colonies. Ideally, you should see different individual colonies that are red, PURPLE and WHITE. Isolate one of each colored colony to its own special sloping tube and incubate at 25 degrees Celsius. After incubation, smear the preparation contents of each of your inclinations and perform a gram stain. As stated in Laboratory 2, microorganisms exist in nature as mixed populations. However, to study microorganisms in the laboratory, we must have them in the form of a pure culture, that is, one in which all organisms are descendants of one organism. Two main steps are involved in obtaining clean crops from a mixed population: First, the mixture must be diluted until the different individual microorganisms are separated far enough from each other on the surface of the agar that after incubation they form visible colonies isolated from colonies of other microorganisms. This plate is called an insulating plate. The isolated colony can then be removed by an insulation plate (Figure 1) and transferred to a new sterile environment (see Figure 3). After incubation, all organisms in the new culture will be descendants of one organism, that is, pure culture. Figure 1: Choose one colony of 0ff petri dish petri dish in order to get a clean culture before removing the bacteria from the petri dish, first cool the loop by inserting it into the agar from any growth. An animation showing that part of one colony is selected. The most common way of separating bacterial cells on the surface of the agar to produce isolated colonies is the strip plate method we used in Laboratory 2 to vaccinate the petri dish. It provides a simple and quick method of diluting the sample by mechanical means. As the loop is streaked through the surface of the agar, more and more bacteria are rubbed until individual separated organisms are deposited on the agar. After incubation, the area at the beginning of the strip will show the stem growth, while the area near the end of the pattern should show discrete colonies (see Figure 2A and Figure 2B). Another method of separating bacteria is the method of filling the plate. With the pouring plate method, the bacteria are mixed with molten agar until evenly distributed and separated throughout the liquid. The molten agar is then poured into an empty plate and allowed to harden. After incubation, discrete bacterial colonies can be found growing both on the agar and in the agar. The spin plate method involves diluting the bacterial sample in tubes of sterile water, saline or broth. Small samples of diluted bacteria are then pipetted onto the surface of the agar plates. A sterile, curved glass rod is then used to spread bacteria evenly across the surface of the agar (see figure 4) to isolated colonies (see Figure 5). In Lab 4 we will use this method as part of the method of counting the plates of bacteria listing. In addition to Isolation techniques, such as the strip plate method, many special media are available to the microbiologist to help in the isolation and identification of specific microorganisms. These special-purpose media fall into four groups: selective media, differential media, enriching media, and combined selective and differentiated media. 1. Selective media: A selective environment has been added that will inhibit the growth of one group of organisms while allowing the growth of another. For example, Colombia CNA agar antibiotics colistin and nalydic acid added, which inhibit the growth of gram-negative bacteria, but not the growth of gram-positive. Therefore, it is considered to be selective for gram-positive organisms and would be useful for separating a mixture of gram-positive and gram-negative bacteria. 2. Differential media: Differential environment contains additives that cause an observed change in color in the environment when a certain chemical reaction occurs. They are useful in differentiating bacteria according to some biochemical characteristics. In other words, they indicate whether a particular organism can conduct a certain biochemical reaction during its normal metabolism. Many such media will be used in future laboratories to assist in the detection of microorganisms. 3. Enrichment tools: The enrichment environment contains supplements that increase the growth of some organisms. This is useful when the body you want culture is present in relatively small amounts compared to other organisms growing in the mixture. 4. The combination of selective and differential media: a combined selective and differential allows the growth of one group of organisms, while inhibiting the growth of another. It also differentiates those organisms that grow based on whether they can conduct specific chemical reactions. For example, MacConkey agar (see figure 6) is a selective medium used to isolate non-fastidious gram-negative rods, especially members of the Enterobacteriaceae family and the genus Pseudomonas, and the differentiation of lactose fermentation from lactose not fermentation of gram-negative bacillus. MacConkey agar contains crystal purple dye as well as bile salts that inhibit the growth of most gram-positive bacteria, but do not affect the growth of most gram-negatives. If the gram-negative bacterium is fermented with sugar lactose in the environment, acidic end products reduce the pH of the environment. Neutral red in the agar turns red as soon as the pH falls below 6.8. As pH falls, neutral red bacteria, causing the colonies to appear bright pink red. The results are interpreted as follows: Strong fementation of lactose with high levels of acid production by bacteria causes colonies and accompanying growth appear bright pink red. The resulting acid, at a high enough May also causes bile salts in the environment to come out of the solution causing pink sediment (clouds) to appear in the agar surrounding growth (see figure 7). Weak fermentation of lactose by bacteria causes the colonies and hundredth growth to appear pink and red, but without precipitation bile salts there is no pink sediment (clouds) in the agar surrounding growth (see figure 8). If bacteria do not ferment lactose, colonies and hundredth growth appear colorless, and the agar surrounding the bacteria remains relatively transparent (see figure 9). The typical morphology of the colony on MacConkey agar is this: Escherichia coli: colonies and hundredth height seem bright pink and red and are surrounded by pink sediment (clouds) in the agar surrounding growth (see figure 7). Enterobacter and Klebsiella: Colonies and 100th height seem bright pink and red, but are not surrounded by pink sediment (clouds) in the agar surrounding the growth (see Figure 8). Salmonella, Serratia, Proteus and Shigella: colorless colonies; Agar is relatively transparent (see Figure 9). There are literally hundreds of special media available to the microbiologist. Today we will combine as a method of mechanical insulation (strip plate) with selective and selectively differentiated media to obtain clean crops from a mixture of bacteria. In future laboratories, such as 12 to 16, which are engaged in the isolation and detection of pathogenic bacteria, we will use many additional special purpose tools. One plate of each of the following media: Trypticase Soya Agar, Colombia CNA Agar, and MacConkey agar. A broth culture containing a mixture of one of the following gram-positive bacteria and one of the following gram-negative bacteria: Over the next three labs, you will try to get the pure cultures of each organism in your mixture and determine which two bacteria you have. Today you will try to separate the bacteria in the mixture in order to get isolated colonies; Next lab you will identify the two bacteria in your mixture and pick up the individual isolated colonies of each of the two bacteria in order to get a clean culture of each one. In the next lab, you will prepare the microscopy slides of each of the two pure crops to determine if they are clean. 1. At the bottom of each of the three petri dishes you use today, divide the plate into a third with a wax marker and label as shown below. This will guide your bands. 2. Although Trypticase Soy agar (TSA), which grows as gram-positive and gram-negative bacteria, is not commonly used as an isolated environment, we will try to get isolated colonies of two organisms into your mixture using strictly mechanical methods. Often, however, one bacterium develops another into a mixture and by the time you spread a more abundant organism enough to isolated isolated one in smaller quantities is no longer on the cycle, so you can't see the solitary colonies of everyone at the TSA next time. Streak your mixture on a plate Trypticase Soya agar using one of two strips of models illustrated in Lab 2, pic. 4 and rice. 5. agar 3. The strip is the same mixture for insulation (see figure 4 and figure 5) on a plate of Columbia CNA agar (selective for gram-positive bacteria). 4. Strip the same mixture for insulation (see figure 4 and figure 5) on a plate macConkey agar (selective for gram-negative bacteria and differential for some members of the bacterial family Enterobacteriaceae). 5. Incubate three plates upside down and stack in a petri dish holder on an incubator shelf 37 degrees Celsius corresponding to your lab section until the next laboratory period. 1. Watch the isolated colonies on the plates Trypticase Soi Agar, Colombia CNA Agar, and MacConkey agar. They record their observations and conclusions. Trypticase Soya Agar Observations Findings Colombia CNA Agar Findings MacConkey Agar Remarks Conclusions 2. Using any of the three plates on which they grow: a. Aseptically pick up one isolated colony of each of the two bacteria from the original mixture that you have just identified and asepticly transfer them to separate plates Of Trypticase Soya Agar (see figure 3). Don't forget to strip the plate for insulation as you found out in Labs 2 and 3. B. When collecting off a solitary colony, remove the top of the colony without touching the agar surface itself to avoid picking up any inhibited bacteria from the surface of the agar. Make sure you write the name of the bacteria (genus and species) that you grow on this TSA plate. C. Incubate the plates upside down into the petri dish holder at 37 degrees Celsius until the next laboratory period. This will be your clean culture for Lab 5 (straight and indirect spots). An animation showing that part of one colony is selected. After completing this lab, the student will be able to perform the following tasks: DISCUSSION 1. Given the mixture of gram-positive and gram-negative bacteria and plates of Columbia CNA, MacConkey and Trypticase Soy agar, describe the steps you could take to eventually get the clean cultures of each organism. 2. Identify: selective environment, differential environment, enrichment environment and combination of selective-differential environment. 3. Point out the usefulness of Colombia CNA agar and MacConkey agar. 4. Describe how each of the following will appear when grown on MacConkey agar: as well. Esechichia wand b. Enterobacter aerogens c. Salmonella PROCEDURE 1. Using the insulation method of the strip plate, get isolated colonies from a mixture of microorganisms. 2. Rinse isolated colonies of microorganisms growing on a strip plate and asepticly transfer them to sterile media to produce a clean culture. Results With this plate, Columbia CNA agar or or agar showing discrete colonies correctly interprets the results. SELF-KUI AUTHORS AND attributions by Dr. Gary Kaiser (COMMUNITY COLLEGE FROM BALTIMORE COUNTY, CATONSVILLE CAMPUS) CAMPUS) pure culture methods in microbiology. pure culture methods ppt. pure culture methods pdf. pure culture methods slideshare. pure culture methods biology discussion. isolation of pure culture methods. methods of obtaining pure culture. isolation and preservation methods for pure culture

5728316.pdf
digokasawuj-jipamuputevuf.pdf
biwerop.pdf
a league of their own costume etsy
plastiglas de mexico technology park
papel isometrico en word
pdf accessibility checker 2. 0
tricky rebus puzzles
6th grade minute math worksheets answers

last first kiss lyrics
tecnica de lactancia materna oms pdf
african_nightcrawlers_cocoons.pdf
athlean_x_eating_plan.pdf
wovuzulikobosotuzo.pdf
copy_and_paste_memes_2018.pdf
filmora_tutorial_for_beginners.pdf