

# Serial dilutions essay



**ASSIGN  
BUSTER**

Many applications require the determination of microbial numbers. Those applications can be either clinical or in a research setting. Clinical applications include determination of antibiotic efficacy and as well as therapy. Research applications include determination of the effectiveness of antimicrobial chemicals, radiation, etc. The viable count is most common or standard method used to quantitate bacteria. With this method only microbes that are alive and able to reproduce can be counted. Since microbes grow into such large numbers it is extremely difficult to perform this without diluting the microbial culture first.

A key concept here is the ability of microbes such as bacteria and fungi to form visible colonies when grown on solid media. This can also be used to count viruses but instead they form plaques on cell culture. A microbial colony is defined as a group of genetically identical cells that originated from one cell on solid media. From that definition we can deduce that in order to grow an isolated colony, one cell should be placed on the solid medium. This also means that one colony reflects the existence of one cell and so can be used to count how many cells were placed on the medium. E. g. f there are 20 colonies on a plate then you must have placed 20 cells to create these 20 colonies.

These cells are therefore referred to as colony forming units or CFUs for short. From the number of colonies and the dilution of the growth on a plate we are able to count the number of microorganism in an original culture. The equation for that: Concentration of culture in CFU/ml = number of colonies on a plate multiplied by the serial dilution on that same plate In this exercise you were browsing through a refrigerator in a microbiology lab and you

found a tube with a solution labeled " B. subtilis:  $2 \times 10^8$  cfu/ml". Now being the skeptic professional that you are, you want to double check the accuracy of the concentration of this culture. Describe how you would embark on this mission, how would you set up the dilutions, which plates would you chose, etc. I would take test tubes of different concentrations of the bacterial samples put one sample of each on a petri dish; diluting the large amount of bacteria to different concentrations. A dilution of  $200 \times 10^{-6} = 2 \times 10^8$  cfu/ml.

You would go about finding this out by taking four other test tubes adding 1.0 ml of to one of the test tubes and shaking them. I would use a general wide series of dilutions ranging from  $10^2$  to  $10^8$ . After shaking this test tube #1 with  $10^8$  dilution, you would take a pipet and transfer 1.0 ml aseptically to the next test tube of  $10^6$ . Shake the test tube of  $10^6$  dilution and aseptically transfer 1.0 ml to the test tube of  $10^4$  dilutions. Shake the test tube of  $10^4$  dilution and aseptically transfer 1. ml to the test tube of  $10^2$  dilution with a pipet. Now shake the test tube of  $10^8$  and take a new pipet transfer 1.0 ml to one petri dish and 0.1 ml to another petri dish. Do this same task to test tubes  $10^6$  and  $10^4$ . I would then go about mixing a agar with the  $10^8$ . I would do this to the remaining plates and allow the agar time to cool and harden. When the plates have hardened I would then go about counting the plates that have between 25-250 colonies. This how I would go about double checking the accuracy of the concentration of this culture.