

Microbiology laboratory questions

[Science](#), [Biology](#)



Review Sheet Week 8 Exercise 21. 5 Questions Differentiate the microscopic morphology of streptococci and pneumococci as seen by Gram stain.

Answer 1. Streptococci are seen as Gram-positive spherical cocci occurring in chains of varying length. Very young cultures may be capsulated.

Pneumococci are observed as Gram-positive cocci occurring in pairs or in very short chains. They are called diplococci as they occur predominantly in pairs. All freshly isolated strains are capsulated. (Ross, 1999. p 264)

2. What type of hemolysis is produced by *S. pneumoniae*?

Answer 2. *Streptococcus pneumoniae* produces narrow zones of α (alpha) or partial hemolysis on blood agar plates. The zones of partial hemolysis appear greenish in color. (Ross, 1999. p276)

3. How is *S. pneumoniae* distinguished from other streptococci with the same hemolytic properties?

Answer 3. *Streptococcus pneumoniae* shares its alpha hemolytic properties with the viridans group of streptococci. However, they differ in other properties. *S. pneumoniae* is optochin sensitive, bile soluble and capsulated. It ferments inulin and produces virulence in mice. While viridans streptococci are optochin resistant, bile insoluble and non-capsulated. They do not ferment inulin and do not produce virulence in mice. (Ross, 1999. p 276)

4. What is the quellung reaction?

Answer 4. The Quellung reaction was first described by Neufeld (1902). The word “ Quellung” means swelling in German. The polysaccharide antigen in the bacterial capsules reacts with the specific antibody produced in sensitized laboratory animals. This reaction causes the capsule to enlarge and become opaque. This can then be observed microscopically. Several

bacterial species namely, *Streptococcus pneumoniae*, *Klebsiella pneumoniae*, *Neisseria meningitidis*, *Haemophilus influenzae*, *Salmonella* and Group B *Streptococci* show a positive test for this reaction. (Fischer et al.)

5. What role does a bacterial capsule play in infection?

Answer 5. The bacterial capsule plays a vital role in virulence and pathogenesis. It protects the bacterium against the host immune processes like phagocytosis. Also, the capsule provides sites for cellular attachment to the host (Hammerschmidt, 2005).

6. What kind of culture media and atmospheric and incubation conditions are best for cultivating streptococci?

Answer 6. Streptococci grow best on nutrient agar enriched with 5-10% blood, heated blood or serum. The optimum temperature for growth is 37°C though growth can occur between the temperature range of 22-42°C. *S. pneumoniae* grows optimally in air or hydrogen with 5-10% CO₂. Incubation period is 24 hours (Ross, 1999. p 264)

7. Why is blood agar considered a differential medium?

Answer 7. Blood agar may be defined both as an enriched and differential medium used for culture of fastidious organisms. A differential medium enables to distinguish one microorganism from another based on the growth properties on the same medium. Blood agar differentiates microorganisms based on their hemolytic properties. Hemolysis is of three types alpha or partial, beta or complete and gamma or no hemolysis. A good example of use of blood agar as a differential medium is seen in grouping Streptococci. *S. pneumoniae* and viridans streptococci are alpha hemolytic, Group A *S.*

pyogenes and Group B *S. agalactiae* are beta hemolytic and Enterococci are gamma hemolytic (Washington, 1996).

8. What is the function of a candle jar?

Answer 8. The candle jar utilizes an airtight jar in which a lighted candle is placed in order to provide an environment rich in carbon dioxide and poor in oxygen. The burning candle utilizes the oxygen in the jar and creates an anaerobic environment suitable for the cultivation of microaerophiles and obligate anaerobes (Collee, 1999. p 117)

9. Describe the hemolysis produced by alpha-hemolytic, beta-hemolytic, and nonhemolytic streptococci.

Answer 9. The alpha hemolytic species of streptococci include *S. pneumoniae* and viridans streptococci including *S. mutans* and *S. sanguis*. The hemolysis produced by these organisms is partial. The hemolytic zone appears greenish in color. The beta hemolytic species are further grouped using Lancefield classification and include Group A species like *S. pyogenes* and Group B species *S. agalactiae*. These species produce complete hemolysis and produce a clear hemolytic zone. The non-hemolytic streptococci are the enterococci species including *E. faecalis* and *E. faecium*. They are the gamma hemolytic species and produce no zone of hemolysis (Ross, 1999. p 264).

10. What type of hemolysis is displayed by the groupable streptococci that are most pathogenic for human beings? To what serological groups do these usually belong?

How can they be identified as belonging to this group without doing a serological test? Explain.

Answer 10. The streptococci that are most pathogenic to human predominantly display beta or complete hemolysis.

Based on the carbohydrate composition of the cell wall antigens they are classified into 20 groups using serological classification proposed by Lancefield (1933). However, the major human pathogens belong to Group A. Group A Streptococci can be also be identified using latex agglutination test, co-agglutination tests and enzyme immuno-assay procedures (Khan).

11. Describe the principle of the latex agglutination test.

Answer 11. The latex agglutination test is an antigen-antibody reaction that is used to detect antibodies to a specific antigen. It is also known as latex fixation test and involves coating of antibodies onto latex particles. The presence of antigen in any sample is detected by visible clumping of the sample. (Livingston, 1991)

12. Name at least three bacterial species found among the normal flora of the throat.

Answer 12. Streptococcus species including *S. mutans* and *S. salivarius*, Staphylococcus species, Lactobacillus species, Corynebacterium species and Neisseria species are commonly seen as the normal flora of the throat (Todar).

13. Is the normal flora of the upper respiratory tract harmful to the human host? Explain.

Answer 13. The upper respiratory tract is often colonized by a large number of organisms in contrast to the lower respiratory tract, which is sterile. Often, many of the organisms that constitute the normal flora may turn opportunistic pathogens in compromised hosts. The upper respiratory tract is

often the initial site of colonization of pathogens and is seen by many as the first region for attack by the opportunistic pathogens (Davis, 1996).

14. Is the normal flora beneficial to the host? Explain.

Answer 14. Normal flora in humans can exist in three states, as beneficial to the host, as harmful to the host and as commensals. Studies suggest that the normal flora influences physiology of the host and also the susceptibility to pathogens. However, the beneficial effects of the normal flora can be seen in the intestines where they participate in bile acid conversion and production of vitamin K (Davis, 1996).

15. In collecting a throat culture, why is it important not to touch the swab to other surfaces in the mouth?

Answer 15. It is important to not to touch the other surfaces of the mouth while taking a throat swab, as the oral cavity is heavily colonized with normal flora. Any contamination by touching these surfaces may give false results for the culture test (Collee, 1999. p 96).

16. What specimens are of value in making a laboratory diagnosis of bacterial pneumonia? Why? Explain the difference between saliva and sputum?

Answer 16. The specimens commonly used in the laboratory diagnosis of bacterial pneumonia are sputum, lung aspirate, pleural fluid, cerebrospinal fluid, urine and blood. Sputum is the most common specimen utilized. Since sputum is expectorated from the lower respiratory tract, it contains the microorganisms that are the actual causative agents of pneumonia. Sputum is the mucus that is expectorated out from the lower respiratory tract while saliva is the watery substance that is secreted by the salivary glands in the

mouth. (Ross, 1999, p. 277)

17. Would a direct Gram stain of a sputum specimen be of any immediate value to the physician in choosing treatment for a patient with pneumonia?

Explain.

Answer 17. Gram stain provides vital information about the organism present in the sample. In case of pneumonia, Gram stain enables preliminary identification of pneumococci as Gram-positive diplococci and hence may be of immediate value to the physician in choosing the treatment (Ross, 1999, p. 277).

18. Does antimicrobial therapy have any effect on the body's normal flora?

Explain.

Answer 18. Antibiotics usually have an adverse effect on the normal flora of the body. Many of the organisms may be wiped out especially those that colonize the intestines (Davis, 1996).

19. What is the significance of VRE?

Answer 19. Vancomycin Resistant Enterococci are species of Enterococcus that have become resistant to the drug vancomycin. This resistance is often acquired through plasmids or transposons. Healthy individuals who have acquired it through nosocomial infections often carry VRE strains for a considerable period of time. The resistance to vancomycin poses a big challenge as the drug is often used as the first line of treatment against enterococci. Further, VRE strains spread rapidly from the healthy carriers to newer hosts (Courvalin, 2006).

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