A BRIEF SURVEY OF THE COCCI

Bacteria that are cocci may exist as benign saprotrophs or virulent pathogens. Many of the saprotrophs are opportunistic pathogens. Inidvidual cells are spherical, but may grow in various cellular arrangements (Fig. 1). In this exercise, students will survey a group of selected species of cocci, some of which are encountered in the environment or in the clinical setting. Others are incorporated in these exercises to support the principles being studied. The intent of this exercise is not to examine all the different species of cocci, but instead to introduce the student to this morphological group of bacteria and how they can be distinguished from one another based upon various physiological properties.

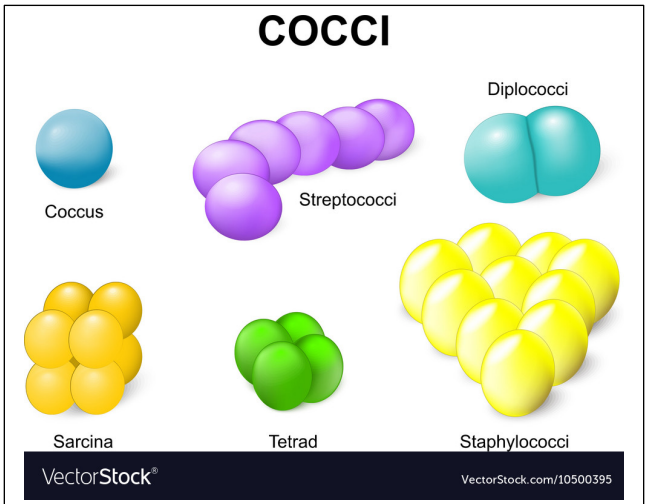

Figure 1. Different arrangements of cocci.

This exercise shall focus on selected members of four Gram-positive groups of cocci, namely species of *Staphylococcus*, *Streptococcus*, *Enterococcus*, and *Micrococcus*. In addition, students will examine the Gram-negative coccus, *Neiserria*. A very brief description of these bacteria is presented here.

Streptococcus: The streptococci are Gram-positive bacteria that tend to form pairs or chains of cells (Fig. 1). The chains may appear bent or twisted. At a rudimentary level, species of *Streptococcus* can be differentiated based upon their ability to lyse red blood cells (RBCs). This property is termed hemolysis. Alpha (α)-hemolytic species partially lyse RBCs and oxidize the iron within them, thereby imparting a greenish color on blood agar media. Beta (β)-hemolytic species will completely rupture RBCs on blood agar plates generating wide, clear areas surrounding bacterial colonies. Gamma (γ)-hemolytic species do not hemolyze RBCs and no greening or clearing of the medium occurs.

α-hemolytic species can be separated into two subgroups: pneumococci and viridans. The former includes the virulent pathogen, *Streptococcus pneumoniae* (which was the agent initially attributed to the death of Kermit's father, but later it was discovered that his death resulted from an infection by *Streptococcus pyogenes*. (Fig. 2; https://en.wikipedia.org/wiki/Jim_Henson). The viridans group, which includes *Streptococcus mutans*, tends to be comprised of relatively benign commensal bacteria that are often associated with the oral cavity.

β-hemolytic streptococci, a number of which are pathogenic, can be further classified by a serological process (Lancefield grouping). There are 20 such described serotypes designated A to V (but excluding I and J). Of particular interest are Group A and B streptococci. *Streptococcus pyogenes* is a virulent Group A pathogen that causes "strept throat" and the pathologies that follow incomplete treatment of infection by this bacterium. Certain strains of *Strept. pyogenes* also causes impetigo, necrotizing fasciitis, pneumonia, scarlet fever, bacteremia, and toxic shock syndrome. Group B streptococci includes *Streptococcus agalactiae*, a known

causative agent of neonatal meningitis and sepsis. Some members of serotype Group D, which includes some opportunistic pathogens, have been removed from the genus and are now characterized as *Enterococcus* or *Lactococcus*.

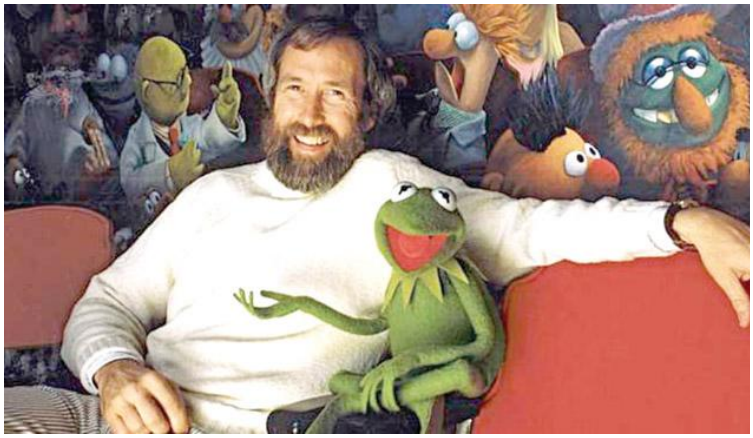

Figure 2. Jim Henson, Kermit, and Various Muppets. (http://www.jordantimes.com/news/features/new-york-museum-honours-kermit-frog-and-his-creator)

Staphylococcus: Staphylococci tend to form clusters of cells that have been previously described as 'grape-like', hence, the term 'staphylo' which in Greek means 'cluster of grapes' (Fig. 1). *Staphylococcus* are Gram-positive and includes more than 40 species. Most are harmless and reside as normal flora in humans and other organisms. They are readily isolated from the soil.

Staphylococcal species can be differentiated from other Gram-positive cocci by several simple tests. The staphylococci are facultative anaerobes (i.e., these bacteria can grow under both aerobic and anaerobic conditions). All species are able grow in the presence of bile salts as well as in media containing 6.5% NaCl (e.g., mannitol salt agar). In addition, *Staphylococcus* species are resistant to the drug bacitracin. Other traits shall be explored in some of the exercises detailed below that distinguish between selected species of *Staphylococcus*.

Among the notable species of *Staphylococcus* are *Staph. aureus*, *Staph. epidermidis*, and *Staph. saprophyticus*. *Staphylococcus aureus* is a member of the normal body flora where it is often found in the nose and respiratory tract as well as on the skin. Most strains of *S. aureus* are opportunistic pathogens causing skin and respiratory infection, sinusitis, and food poisoning. The emergence of antibiotic-resistant strains, such as methicillin-resistant *S. aureus* (MRSA), is a worldwide problem. A vast majority of *Staph. aureus* strains are β-hemolytic and possess the enzyme coagulase as a virulence factor (described below). In contrast, *Staph. epidermidis* is α-hemolytic, does not possess coagulase, and is considered generally benign. It is part of the normal human flora, but immune compromised patients are at risk of developing infection by this bacterium. Usually, such infections are hospital-acquired especially for individuals having catheters or surgical implants. By comparison to the other two species above, *S. saprophyticus* is also α-hemolytic and coagulase negative. It is usually found as part of the normal flora in the female genital tract. *Staphylococcus saprophyticus* can be differentiated from *Staph. epidermidis* by testing for susceptibility to the antibiotic novobiocin. *Staphylococcus saprophyticus* is resistant to novobiocin, whereas *S. epidermidis* is novobiocin-susceptible.

Enterococcus. The enterococci are a large group of Gram-positive lactic acid bacteria that had previously been difficult to distinguish from the streptococci based solely on physiological characteristics. Two species are common commensals in the intestines of humans and animals – *Enterococcus faecalis* and *E. faecium*. Enterococci are tolerant of a wide range of temperatures (10–45°C), pH (4.5–10.0), and high salt (NaCl) concentrations. These bacteria are typically α-hemolytic on sheep's blood agar. Species of *Enterococcus* cause a variety of infections and often exhibit intrinsic antibiotic resistance. Particularly virulent strains of *Enterococcus* are resistant

to vancomycin and have emerged as a serious cause of nosocomial infections. In addition, the presence *Enterococcus* in drinking or recreational waters generally indicates fecal contamination.

Micrococcus. Species of *Micrococcus* occur in a wide range of environments such as soil and water. Micrococci are Gram positive and can be observed as tetrads or as sarcinae (Fig. 1). Some species are pigmented, such as *M. luteus* (yellow) and *M. roseus* (pink/red). *Micrococcus* is generally thought to be a saprotroph or commensal microbe. However, it is an opportunistic pathogen in immune compromised hosts. Micrococci are metabolically versatile which has been exploited by industry to produce a variety of microbial compounds including herbicides and chlorinated biphenyls.

Neiserria. The genus *Neiserria* is comprised of 11 Gram-negative species of diplococci (Fig. 1). All colonize humans, but only two are human pathogens – *N. meningitidis* and *N. gonorrhoeae*. The former causes bacterial meningitis and septicemia, whereas the latter species is well known as the agent of the sexually transmitted disease, gonorrhea. The remaining 9 species of *Neiserria* are considered benign although they can occasionally cause disease. Most species of *Neisseria* are positive for both catalase and oxidase activity, but individual species are identified based upon the production of acids from different sugars.

Figure 3. Scanning electron micrographs (false-colored images) of *Staphylococcus aureus* (left image), *Staphylococcus salivarius* (middle image), and *Enterococcus faecalis* (right image). (http://www.sciencephoto.com/set/3193/dennis-kunkel-bacteria-images)