Live Type A Vaccine for *F. Tularensis* for the Southwest United States

Pathogen – *Francisella tularensis*

Vaccine type –Llive Attenuated

Study population – Southwest United States

Literature Review:

Tularemia is a gram-negative bacterial zoonosis, which is caused by Francisella tularensis (F. tularensis). This disease has been identified in all states but Hawaii, with the majority of cases occurring in the south central and western regions of the United States (Dennis 2001). Tularemia causes sudden fever, chills, headaches, diarrhea, muscle aches, joint pain, ulcers on hands or mouth, often death and these symptoms generally become visible in 1-14 days prior to infection (CDC 3). The infection is caused usually during rabbit hunting season because rabbits act as the reservoir of this disease and it is transmitted to humans by ticks and deer flies that act as the vector for infection. Another method of infection may be from ingesting infected soil, water, or food. Inhalation of this disease is sometimes caused during the skinning of rabbits or more importantly used as an airborne agent in a biochemical attack from terrorist organization (CDC 2).

Tularemia is an intracellular bacterium that invades and replicates through macrophages. It is a nonmotile, aerobic, gram-negative coccobacillus and this stimulates the making of inflammatory cytokines (Loegering 1).

The antibiotics used in treatment of Tularemia are mostly still experimental and not approved by the FDA(Food and Drug Administration), but the most commonly used

treatment for an infection is streptomycin. Others include gentamicin, tetracycline, and chloramphenicol (Dennis 2001).

Preventions to Tularemia include insect repellent containing DEET to prevent ticks. Another prevention measure includes wearing gloves, and safe handeling of dead rabbit carcasses. Using safe, uninfected water is extremely important as well.

The infection begins at the epithelial level and replication occurs within the body, the first immune response is from macrophages. The macrophage recognizes the antigen from surface receptors and at this time cytokines are released to the body signaling the neutrophils and complement to go to the site. With this disease being gram-negative, complement is not effective in combating the antigen infection.

This disease was chosen because of the national attention to bioterrorism and the ability of this disease to mutate quickly making it incredibly hard to specifically find and lyse. The current treatment options to this disease are another reason to why this was chosen because of the percentage of death in undiagnosed cases. This disease is so under diagnosed because the similarities of the flu and pneumonia symptoms.

Description of Vaccine:

The type of vaccine used will be attenuated live vaccine. The sub-species, tularensis Type A of the live vaccine strain will be used in particular for the Francisella tularensis strain. There are two types of vaccine, Type A and Type B that is currently available for use. The Type B, holarctica, is currently in use and is not as strong as the Type A vaccine strain, therefore the Type A has not been used. The type of immunity in target is the adaptive immunity, which includes the activation of B cells to produce

antibodies and since the Type A strand is stronger the vaccine will be as well. B cells are soluble pathogen-binding molecules of the adaptive immunity that circulate body fluids to allow the binding of antibodies to bacterial cells and intact viral particles in extracellular spaces. This leads to the phagocytosis of the pathogen. To target these pathogens, the binding sites of antibodies interact with intact components of the surface of the antigen, such as glycoproteins and proteoglycans.

Antigens in the tissues are transported by macrophages, PMNs, and dendritic cells to close-by secondary lymphoid organs. This is where the activation of lymphocytes occur to become immune effector cells. T cell lymphocytes arise from stem cells in bone marrow and leave for the thymus to mature. During development in the thymus, clonal selection occurs. Positive selection then happens which saves T cells that have receptors reacting well for self MHC I and MHC II. The other cells are negatively selected and induce apoptosis. T cells that recognize MHC I on dendritic cells (CD8 T cells) go to pathogen and kill infected cells. Adaptive effector functions increase for about 10-14 days after presentation to antigen, they plateau for several days, and then drop once antigen has been eliminated. Yet even at this dropped level, the effector functions are at higher concentrations. The main antibody produced during this first response is IgM followed by IgG. Upon the next event of the antigen contact the lag period is much briefer, the peak responses are higher, and the antibody levels are increased for weeks or months. IgG, as well as IgA, are the main antibodies for secondary immune responses.

The reason for choosing this pathogen is due to the non-effective vaccine currently in use and is a biological warfare threat; therefore it is important to have a defense against this sort of weapon. The live attenuated vaccine because the pathogen

mutates at a fast rate and also because it shows the most progress in creating immunity against the antigens. The problems that could arise are side-effects due to the live vaccine and any developments of vaccine resistance against such a strong strain.

The current vaccine is not always effective because it does not always detect the lipoproteins on the outside membrane of F.tularensis. This organism lives within vacuoles of infected macrophage cells. The THP-1 cytokines are stimulated to make the CD8 put holes in the cells by using perforin, and then this causes the pathogen to become extracellular. Once extracellular, dendritic cells present it and allow for more T cells to be produced.

Description of Immunity Assessment:

The sample pool consists of 100 adults in the age range of 18-60 year olds. The subjects will be vaccinated and tested for specific antibodies at 1 week, 2 weeks, 1 month, and 3 months. The test that will be used to measure antibodies is the Elisa assay. The T cell responses can be measured by cytotoxicity. The circulating T cells will be separated from erythrocytes and other leukocytes, in which they will be exposed to antigen shown on self-class I or II on MHC to recognize T cell activation. Target cell lysis will be detected by inclusion of 51 Chromium. Secreted cytokines (THP-1) will be detected by binding them to their specific antibodies.

A possible problem to our immunity assessment is funding. This can occur by a powerful resistance from Tularemia strain to such a strong vaccine. By having a strong vaccine produced, any resistance to this would result in the need of producing a higher and stronger vaccine. This requires more government funding for the research and production, which is hard to achieve t times. Also, this causes aims at controlling and

responding to a vaccine resistance will require a large budget that could cause deficit by government funding, if any is given. The proposed vaccine is expected to be successful because type A, the type that will be used, is a stronger species than Type B the species that was used in previous vaccines. Also, success is expected because a live attenuated vaccine will be used rather than a killed vaccine used in previous vaccines. This live attenuated vaccine is expected to stimulate generation of memory cells as well as humoral immune responses. Because the ability of the virus to multiply within the host, less virus will be required to be injected to induce protection. By using such a strong strain, the side effects may be more severe. Secondary infection is another safety issue. With this stronger vaccine a mutation causing a stronger strain could occur. In the case that this is picked up by a secondary vector, a stronger outbreak could occur. By using the whole virus, the stimulation of response to antigens in their natural conformation will be allowed. Also, live attenuated can be administered orally so the cost of its utilization will be lower. Also, oral administration allows for mucosal immunity and IgA synthesis. This gives more protection at the normal area of virus entry which is where the tick bites the victim.

Information Sources:

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Frequently Asked Questions (FAQ) About Tularemia.
<http://www.bt.cdc.gov/agent/tularemia/faq.asp>