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CLINICAL INTERNSHIP WITH THE PEDIATRIC CLINIC'S CLINICAL RESEARCH AT THE PATIENT CARE CENTER OF THE UNIVERSITY OF NORTH TEXAS HEALTH SCIENCE CENTER/ TEXAS COLLEGE OF OSTEOPATHIC MEDICINE: LITERATURE REVIEW OF MENINGOCOCCAL MENINGITIS

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THESIS

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By

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THESIS TOPIC

Epidemic meningococcal meningitis and meningococcemia disease is caused by the bacterial pathogen *Neisseria meningitidis*. Once infected with meningococci, onset of the disease is rapid with a high rate of morbidity and mortality. Without medical intervention the mortality rate is over 50%. Medical treatment of an outbreak of the disease with antibiotics can reduce the death rate to 10-15%. However, 10-20% of survivors will suffer from neurological damage that may include loss of hearing, paralysis or mental retardation. Recent concerns have been noted regarding the emergence of Neisseria meningitidis strains resistant to antibiotics.

Vaccines have been developed in an effort to reduce epidemic outbreaks of meningococcal meningitis and meningococcemia. The first generation polysaccharide vaccines have shown to be safe and possess some degree of effectiveness but have shortcomings of limited length of immune protection and evidence of hyporesponsiveness to subsequent vaccinations. The second generation conjugated polysaccharide vaccines have been able to overcome these problems and show great promise in reducing the scale of epidemic meningococcal outbreaks with implementation of effective mass vaccination campaigns. In addition, reducing the number of infections will limit the exposure of *Neisseria meningitidis* to antibiotics and, in theory, slow the development of resistance to antibiotics.

INTRODUCTION TO BACTERIAL MENINGITIS WITH EMPHASIS ON INFECTION WITH *NEISSERIA MENINGITIDIS*

Only the gram-negative bacterial organism Neisseria meningitidis causes epidemic meningococcal meningitis and meningococcemia (12). Meningococcal meningitis is the most common syndrome [CNS] and refers to an infection by *N. meningitidis* to the central nervous system with accompanying inflammation of the meninges surrounding the CNS. The typical signs and symptoms of meningococcal meningitis are fever, headache and a stiff neck. In children less than one year of age, a bulging fontanelle (soft spot on the head) is suggestive of meningitis. A lumbar puncture usually reveals am abnormal, cloudy cerebrospinal fluid (12). Meningococcemia is characterized by an infection by the bacterium *N. meningitidis* that causes septicemia, or an infection of the bacteria in the bloodstream. Meningococcemia is less common but considered more severe and can present alone or with meningococcal meningitis. The common symptoms of septicemia are fever, petechial or purpural rash which are small areas of bleeding in the skin, and low blood pressure. Some patients have seizures, coma, or other evidence of altered mental status. During epidemics 10-20% of patients present with septicemia. A lumbar puncture may reveal cloudy or a normal appearing clear water-like cerebrospinal fluid (13). Both meningococcal meningitis and meningococcemia are diseases that progress rapidly with a high degree of morbidity and mortality (12).

The primary site of infection normally occurs in the nasal and pharyngeal areas. The bacteria are found on the mucosa of the nose and throat and spread from person-to-person in oral secretions or respiratory secretions (12). In a population where there is an outbreak as many as one in ten people are unaffected carriers of Neisseria meningitidis (30). It is not currently known why some people are unaffected carriers of N. meningitidis, but humoral immunity probably plays a role (12).

Epidemic meningococcal meningitis is largely seen in sub-Saharan continental Africa and in developing nations of other continents (29). In the African 'meningitis belt' periodic epidemics begin in December, the dry season, and end in May, the rainy season (12). The reasoning for this periodic seasonality in Africa will be discussed in the epidemiology review. Large outbreaks of meningococcal meningitis and meningococcemia did occur in the nineteenth and earlier half of the twentieth centuries (24) in the United States. The diseases are now mainly seen in situations where groups of individuals live in close proximity, as seen in the sporadic outbreaks occurring in the military and amongst college freshman living in dormitories (29).

Treatment of the diseased state can be successful when diagnosed early, but extensive damage often cannot be averted. However, even with medical intervention, the mortality rate is around 10%. Of those who survive, 10% suffer from neurological complications such as loss of hearing or paralysis (28). Other bacterial and viral organisms may cause meningitis, the swelling of the meninges around the brain and spinal cord, but are not associated with meningitis epidemics.

LITERATURE REVIEW

Historical Background of Bacterial Meningitis

Recognition of the Symptoms of Bacterial Meningitis

Bacterial meningitis has a long history in man as described in a Mesopotamian incantation dated from around 4000 to 3000 BC. The incantation describes the region of epidemic prevalence within the sub-Saharan Africa meningitis belt, along with symptoms of headache and nearly certain forthcoming death (24).

Hippocrates described the association of headache and tinnitus with inflammation of the brain, including recognition of the high mortality associated with these symptoms of bacterial meningitis. In 460 BC, Hippocrates described a syndrome of purulent otitic fever associated with cerebral symptoms as follows: "We need to pay attention in acute ear pain accompanied by fever because the patient can become delirious and, in a short time, die" (24). However this description has been attributed to a case of brain abscess in association with otitis. Hippocrates may have believed that the intracranial infection was primary and that involvement of the ear was secondary, with the ear being a conduit for drainage of focal suppuration of the brain (24).

The first clear account of meningococcal meningitis is usually credited to Vieusseux, a Swiss physician who described a small outbreak of meningitis in Geneva in March 1805 (19). The symptoms of violent headache, vomiting, stiffness of the spine, and livid patches on the skin were described by Vieusseux in several of the cases (24).

Vladimir Mihailovich Kernig developed a technique for diagnosing suspected cases of meningitis (24). With the patient in a seated position, the neck was flexed anteriorly towards chest. Neck stiffness and rigidity is indicative of a positive sign of possible meningitis (24). Josef Brudzinski, a Polish physician, developed a similar technique, the nape of the neck sign. With the patient lying on their side, flexion of the neck was performed by the examiner, flexion of the hip and knee was interpreted as a positive nape of the neck sign. Early reports by Brudzinski stated that his neck sign was seen in 96% of cases of meningitis versus 57% in Kernig's sign (24).

Isolation of Bacteria from the Cerebrospinal Fluid

The first report of meningococcal isolation was attributed to Anton Weichselbaum in 1887 (24). Weichselbaum isolated gram-negative diplococci bacteria from the CSF of a patient who had died of sporadic meningitis and isolated the organism. He called the bacterium Diplococcus intracellularus meningitidis (24).

Heinrich Quinke developed the technique of lumbar puncture in 1891. The development and perfection of the technique of lumbar puncture by Quinke has made possible the formal examination of cerebrospinal fluid [CSF] and the diagnosis of meningitis (24). Ludwig Lichthein, in 1893, observed that CSF glucose concentrations were low in the presence of bacterial and tuberculous meningitis. His analysis of CSF by biochemical methods is the earliest example of the diagnostic value of the lumbar puncture and CSF analyzation (24). Measurement of CSF glucose concentrations and other assays are now used to confirm the presence of bacterial organism invasions of the central nervous system (2). There are several surveillance techniques used in examining

the CSF, some of which include isolation and identification of organisms, detection of bacterial antigens, elevated leukocyte levels, glucose and lactate concentrations of the CSF, measurement of pH level of CSF, and protein concentration of the CSF. These various diagnostic and measurements of the CSF are crucial in today's approach to diagnosing bacterial meningitis and choosing the correct therapeutic treatment regiment (2).

Historical Methods in the Treatment of Bacterial Meningitis

Prior to the introduction of antisera, bacterial meningitis was a particularly lethal, with a mortality rate ranging from 70-90% (21). Devastating epidemics of meningococcal meningitis occurred in New York in 1904 to 1905 and eastern Germany in 1905 to 1907. Following these epidemics, investigators from these countries were working almost simultaneously on the development of a meningococcal antisera (24). The development of an antiserum against meningococcal disease substantially decreased the mortality rate from 70-90% to 30%-40% (21). Proof of the effectiveness of antiserum therapy was demonstrated in military personnel during World War I. Sixty-seven percent of the nearly 2500 military personnel admitted to hospitals in the United States with meningococcal meningitis survived with the antiserum treatment (21).

Antibacterial activity of sulfonamides was discovered in the early 1930's (20). Research done by Schwentker using sulfonanilamide, a sulfonamide compound, was reported in 1937 (20). His study involved the treatment of 11 patients with confirmed meningococcal meningitis. Out of the 11 patients treated with meningococcal meningitis, 10 survived. The one death had sterile CSF and blood cultures and was contributed to

pneumonia, but may have been attributable to a superinfection, a presence of toxic cellular components in the blood. A mortality rate of only 9% signified a significant advance in therapeutics for meningococcal meningitis and thus, sulfonamides became the first chemotherapeutic drug class against *N. meningitidis*. The successful treatment of the son of President Franklin Roosevelt for a streptococcal throat infection with a sulfonamide in 1936 was probably significant in making sulfonamides widely available to American physicians. However, sulfonamide therapy ended with the emergence of sulfonamide-resistant meningococci in 1965 (21).

Fleming's discovery of penicillin in 1931 was able to fill the gap created with the removal of sulfonamides from use as antibacterial agents against *Neisseria meningitis*. During World War I, a study conducted by Rosenberg and Arling addressed the use of penicillin in 71 U.S. servicemen where the median age was 18 (20). All but one of the patients survived without sequelae [persistent damage or disability caused by the disease]. The one fatality was a patient that was comatose with a temperature of 108° at admission. Despite penicillin's effectiveness, sulfonamides remained the drug of choice until strains of sulfonamide-resistant meningococci began to emerge (20).

Today, penicillin is the drug of choice for the treatment of meningococcal meningitis, however, varying degrees of resistance to penicillin has been seen in some regions of the world. A third generation cephalosporins can be used alone or in combination with vancomycin when resistance may be suspected (24).

The development of bacterial resistance to antimicrobial therapy underscores the need for safe and effective vaccines and vaccination campaigns against *Neisseria meningitidis*.

Successful vaccines may reduce the scale of meningococcal meningitis epidemics. The *Haemophilus influenzae* vaccine is a good example of a vaccine that was developed and through a vaccination campaign in the United States, targeted it's most susceptible population, infants and children, dramatically reducing the incidence (8).

Vaccination campaigns in third-world regions such as the African meningitis belt would be challenging. Limited government resources and limited medical infrastructure combined with large rural populations would make implementation of a vaccination campaign a daunting task. However, with cooperation of the governments and participation by entities such as the United Nation's World Health Organization, a vaccination campaign could theoretically be successful, at least in building herd immunity in a population. Herd immunity is defined as a portion of an unvaccinated population that is protected from disease due to an effective vaccination campaign of the larger population. The vaccination of the larger population reduces the available human reservoir for infection and spread of the disease (1).

Meningococcal Meningitis: *Neisseria meningitidis*

Epidemiology

The five *N. meningitidis* serogroups that are responsible for the majority of the cases of meningitis and meningococcemia are A, B, C, Y and W-135 (15). Serogroup A is predominant in Africa (11) while Serogroup C (29) tends to be more prevalent in Europe and North America. Serogroup B tends to be seen most in temperate climates (15) and serogroups Y and W-135 are more erratic in outbreaks.

The highest burden of disease occurs in sub-Saharan Africa. This is the area between Senegal in the west and Ethiopia in the east, which is referred to as the meningitis belt. In this region epidemics occur in seasonal cycles between the end of November and the end of June, depending on the location and climate of the country. The epidemics decline rapidly with the arrival of the rainy season (30). It is thought that the low humidity and dust may damage normal mucosal barriers in the nose and throat, this allowing N. meningitidis to invade the blood stream, or to be transmitted from person to person more easily (12). Within the meningitis belt, meningococcal disease has traditionally occurred in epidemic cycles, which tend to occur every 8 to 15 years. However, the cycles appeared to have shortened and become more irregular in the past 20 years (27). The mechanisms that cause these cycles are not well understood, but are believed to be related to variations in herd immunity (30). The irregularity of the disease cycles may be attributable to the vaccination campaigns using meningitis A serogroup polysaccharide vaccines that have shown short-lived immunogenicity in children (15) and meningitis C

polysaccharide vaccines that induce a hyporesponsive immune reaction to meningococcal antigen when the vaccine is given in series (15). The hyporesponsive immune reaction to challenge by bacterial antigen failed to produce antibody against meningococcal C to the levels produced prior to vaccination, in effect lowering the immune defense against meningococcal challenge.

The shortened cycles continue in the meningitis belt with little reprieve (25). In 1996, approximately 300,000 cases of meningococcal disease were reported to the World Health Organization in a pandemic that primarily effected Nigeria, Burkina Faso, Niger and Mali, all within the meningitis belt (27). In a two year period, 1988-1989, more than 127,000 cases of meningococcal disease were reported from Africa, with over 40,000 cases reported from Ethiopia in 1989. Outbreaks of meningococcal meningitis occurred outside of the traditional meningitis in several large epidemics in Nairobi, Kenya in 1989, in Tanzania in 1990-1992, and in Burundi in 1992 (12). "The reasons for this unusual pattern of disease are not well understood, but it may be related to a unique group of *Neisseria meningitidis* strains" (12), or variations in herd immunity. Humoral immunity, pre-existing antibody against N. meningitidis, in populations may explain the 8-12 year cycle of epidemic meningococcal disease that has been observed in the meningitis belt (12). Epidemics may not occur until the humoral immunity to a particular strain in a population has declined. Humoral immunity may have been inadvertently shortened with the use of polysaccharide vaccines in infants and children, the segment of population at greatest risk to meningococcal disease.

Epidemics are not isolated to the African continent alone as epidemic meningococcal disease can occur in any part of the world (30). For example, Asia has had some major epidemics of meningococcal disease in the last 30 years including China 1979 and 1980, Viet Nam 1977, Mongolia 1973-1974 and 1994-1995, Saudi Arabia 1987, and Yemen 1988 (30).

The United States annually records approximately 3000 cases of meningococcal disease each year, with a 10-13% mortality rate (31). It is worth noting that even with the sophisticated medical infrastructure present in the United States, mortality rates for meningococcal disease are similar to mortality rates of those treated in third world countries. The *CDC Personnel Health Guideline* states that, serogroups B and C cause 46% and 45% of the endemic cases, respectively (25). Epidemic meningococcal disease has, since the early 1990's, been caused increasingly by serogroup C (31).

The most practical way to control an epidemic of meningococcal disease, according to the CDC's Epidemiology Program Office, is to begin mass vaccination as soon as an epidemic is identified (12). Earlier detection translates to fewer deaths with respect to vaccination campaigns. The number of deaths from epidemic meningococcal disease can be reduced by the following (12):

1. Early recognition of cases and correct case management by health care workers;
2. Encouraging people to seek medical care for symptoms of meningococcal disease;
3. The availability of appropriate antibiotics at health care facilities; and

4. Rapid mass vaccination, which will reduce the number of cases and deaths that occur.

Risk factors identified for contracting meningococcal disease during epidemics have been compiled by the CDC have shown that crowded living conditions and low socioeconomic status have been associated with disease (12). Concurrent upper respiratory infections, nutritional status and malaria are also associated with disease. The possibility of immune deficiency caused by infection with human immunodeficiency virus [HIV] was explored as a risk factor due to the high incidence of the disease in Africa. However, it was found that infection with [HIV] does not appear to be a significant risk factor for serogroup A meningococcal disease during epidemics (12).

The development of conjugated polysaccharide vaccines against *N. meningitidis* looks to be a viable method of creating long lasting immunological memory against serogroups A, C, Y and W-135. The near eradication of *Haemophilus influenzae B* [Hib] due to the success of its conjugated vaccine has spurred similar hopes of such effectiveness of a conjugated vaccine against meningococcal meningitis. Conjugate vaccines are not only effective in adults but also in infants and children (3). Moreover, the conjugated vaccine is able to overcome the problem of hyporesponsiveness in adults (18) as well as in children (3, 8).

A larger numbers of trials should be conducted in Africa's meningitis belt in the interest of Justice described in the Belmont Report (13). This region and its people bear the greatest burden of the disease and as such are entitled to receive the benefits of such research. Complications arise, however, when doing research in countries not covered

under FDA regulations or the International Conference on Harmonization [ICH]. ICH members are the United States, the European Union and Japan (11). ICH observers, such as Canada and the World Health Organization, act as links between non-ICH countries and have chairs on the steering committee. The ICH has guidelines similar to those of the FDA in the United States for managing the process of drug trials in humans. Countries not following regulations or guidelines such as those of the ICH may place human research subjects at significant risk. This lack of guidance and oversight of pharmaceutical manufacturers that perform trials in third-world countries was described in a series of articles titled The Body Hunters in the Washington Post (32). In addition, these articles explore how cultural differences, language barriers, education and rural and migratory demographics further complicate the conduct of clinical trials. These issues combined with lack of regulatory oversight have made informed consent, adequate long term follow-up and the interpretation of clinical data difficult. Nonetheless, it is the third world regions that carry the burden of meningococcal disease. The testing of investigational drugs becomes extremely difficult in regions where regulatory authorities lack jurisdiction to oversee research. As pointed out in the Washington Post series (32), the FDA now requires that any drug being tested in other countries for marketing in the United States must be approved for testing under FDA regulations. This ruling forces pharmaceutical and device manufacturers to create protocols in designing their trials that are acceptable to FDA regulators and creates a paper trail that can be examined.

A future study could look at the cost effectiveness of using a combination of polysaccharide vaccine and conjugated vaccine in series versus the use of the conjugate vaccine alone in series. The hypothesis is that cost would be lower using less expensive polysaccharide vaccines in second and third series injections, while still providing protection and circumventing the hyporesponsiveness (3) seen in meningitis polysaccharide C vaccines alone in serial injection.

Pathogenesis and Pathophysiology of Neisseria meningitidis

Successful colonization and invasion of the host by gram-negative *Neisseria meningitidis* is based on the organism's ability to evade the host's barriers and immune defenses (17). Host factors that are that are favorable to meningococci infection may also play a role in colonization. Upper respiratory infections, alcoholism (14) and smoking (29) have been identified as risk factors for meningococcal disease. Apart from these risk factors, meningococci have several virulence factors that allow them to gain a foothold in the host.

N. meningitidis colonizes in the nasopharynx, but in order to do so it must be able to avoid being swept away by epithelial ciliated cell movement of mucous towards expulsion. The bacteria must also be able to penetrate the thick glycoprotein mucous that coats the epithelia while avoiding secretory IgA antibodies present in the mucous (17). Meningococci secrete IgA proteases that cleave secretory IgA, destroying the antibody's ability to identify the bacteria as foreign to other immune components. In addition, the meningococci are able to locally damage ciliated epithelial cell, causing a loss of the movement of the cilia.

Attachment to the epithelium is mediated through the use of pili that attach the bacteria to non-ciliated cell surface. The non-ciliated epithelial cell engulfs the bacteria endocytotically and transports it in a membrane bound vacuole through the cell to the basolateral surface and releasing the bacteria to the extracellular space (17).

From the extracellular space *N. meningitidis* move to the vasculature to enter the circulation, evading macrophage phagocytosis in the extracellular space by way of its polysaccharide capsule (17). It is also the capsular polysaccharide coat that allows the meningococci to evade phagocytosis by neutrophils and the alternate complement pathway while in the bloodstream (17).

The complement pathway is the chief host defense mechanism against bacteremia (9). The complement system is activated in a cascade mechanism that systematically cleaves inactive zymogens, to activated, complement pathway proteins. The complement system has three recognized pathways that lead to recruitment of phagocytes and inflammation, opsonization which coats the pathogen and targets it for removal, and the formation of membrane attack complexes that form pores in the outer membrane of the pathogen, that can lead to the death of the pathogen (9).

It is the capsular polysaccharide that allows *N. meningitidis* to escape the complement pathway of the immune system (9). The meningococcal polysaccharide capsule contains sialic acid, as do erythrocytes on their plasma membrane. The presence of sialic acid facilitates the binding of the regulatory protein, factor H. Factor H inhibits the activation of the alternate pathway of the complement cascade by binding to complement protein C3b, blocking the first step in the alternate pathway cascade (9, 17).

Meningococci in the bloodstream have receptors that preferentially bind to receptors present on vascular endothelial cells in the choroid plexus region of the central nervous system circulation (19). The choroid plexus is an in-folding of arteries located in the third, fourth and lateral ventricles of the brain (1). Adherence of meningococci to the vascular endothelium is followed by invasion of the bacteria into the subarachnoid space and the cerebrospinal fluid. The subarachnoid space is formed between the dura and pia mater of the meninges (1). The cerebral arteries lie within the space with the CSF being formed by ultrafiltration of the plasma through the vasculature of the choroid plexus. The choroid plexus is an area of the vasculature that differs from vasculature that serves the majority of the CNS. The choroid plexus is somewhat fenestrated, containing small pores, which allows for a greater filtration than that of the tightly closed vascular endothelium, or blood brain barrier, of the rest of the vasculature that serves the CNS (7).

Once the meningococci have entered the cerebrospinal fluid there is little presence of immune response elements due to the blood brain barrier's exclusion of large particles and proteins (10). The bacteria are able to multiply quickly in this environment, indicative of the quickness of the presentation of bacterial meningitis. The presence of meningeal and perivascular macrophages probably play a role in recruiting granulocytes and polymorphonuclear [PMN] leukocytes to the area of infection by release of inflammatory and chemotactic cytokines, tumor necrosis factor-α [TNF-α] and interleukin-1β [IL-1β]. Astrocytes and microglial cells of the CNS have also shown an ability to release cytokines in the presence of bacterial pathogens (10). TNF- α promotes the expression of molecules on the vascular surface of endothelial cells that promote

adherence of neutrophils and induces IL-1 synthesis and release from astrocytes and microglial cells (19). IL-1 has chemotactic influence on leukocytes and stimulates granulocyte neutrophils, a class of leukocyte, to release toxic oxygen metabolites by degranulating (17).

Activated granulocytes, although are the major clearance mechanism in bacterial meningitis also contribute to the deleterious affects of bacterial meningitis with their cytotoxic activities (10). The toxic oxygen bursts released upon degranulation can damage the vascular endothelium and nerves in the CNS (17). Damage to the vascular endothelium results in an increased permeability of the blood brain barrier. With the blood brain barrier compromised, proteinaceous components of the blood can diffuse into the subarachnoid space. Water follows the osmotic gradient into the subarachnoid space and contributes to a rise in intracranial pressure and edema. The CNS loses its ability to effectively remove the excess fluids due to the accumulation of serum proteins and cellular debris. The increased intracranial pressure forces the accumulating fluids into the brain parenchyma. The accumulation of the toxic substance containing fluids in the brain parenchyma can have damaging effects on the brain including cerebral herniation (17).

Release of the meningococcal cellular components, namely the lipopolysaccharide [LPS], causes further release of inflammatory cytokines by leukocytes and vascular endothelial cells (16). The further release of inflammatory and chemotactic cytokines contributes to the removal of the bacteria and cytotoxic components but also causes further damage to the vascular endothelium by the extravasion of more leukocytes to the

site and possible vascular thrombosis, occlusion of vessels due to an accumulation of cellular elements. Damage to the vasculature and LPS can cause decreased blood flow to the CNS and vasospasms that can lead to cerebral ischemia (19).

Extensive damage to the central nervous system and vasculature that supplies it occurs when the immune response elements are activated in bacterial meningitis. The overwhelming response of the immune system to clear the bacteria and LPS capsular components from the CNS causes profound effects and is reflected in the high morbidity and mortality rate seen in meningococcal meningitis. Intervention by treatment with antibiotics can curtail much of the damage if the disease is recognized.

Clinical Presentation of Meningococcal Meningitis

The characteristic symptoms of meningococcal meningitis are sudden onset of intense headache, fever, nausea, vomiting, photophobia and neck stiffness (26). Indications of neurological involvement include lethargy, delirium, coma and/or convulsions. Symptoms in infants may be more subtle and nonspecific but may include fever [50%], lethargy, poor feeding, respiratory distress, irritability, vomiting and diarrhea, seizures [40%] and a bulging fontanelle [30%], the soft spot on the head (6). Presentation in older adults is likewise, more subtle, and may not include the classical symptoms. However, a retrospective study discovered that the one of the symptoms of fever, neck stiffness, or altered mental status was present in virtually all patients with meningitis [sensitivity of 99%-100% for the presence of one of these findings]. Thus, the absence of any of the 3 symptoms essentially excluded the diagnosis. The presence of all 3 findings were not common [pooled sensitivity of 46%] (5).

The clinical presentations of meningococcemia are fever, petechial or purpural rash, which is small areas of bleeding in the skin, and low blood pressure (11). The clinical signs of both meningococcemia and meningococcal meningitis may be present together. Apart from the classical symptoms previously mentioned, use of Kernig's and Brudzinski's signs should be employed in cases of suspected bacterial meningitis.

Lumbar puncture is performed when the clinical presentation suggests the presence of bacterial meningitis. Analysis of the CSF should include glucose and lactate concentrations, white blood cell counts, gram-staining, and protein concentrations. The majority of the cases of bacterial meningitis will have: CSF/serum glucose concentration ratio ≤ 0.40, elevated CSF lactate concentration versus normal lactate concentrations in viral meningitis, CSF pleocytosis of WBCs, a positive gram-staining culture, and elevated levels of protein. Treatment with oral antibiotics prior to obtaining CSF from the lumbar puncture does not significantly alter the ability of the clinician to diagnose bacterial meningitis (2). Statistical analysis has shown that the difference in CSF total WBC count, percent of neutrophils and glucose concentration are not significantly different between untreated and pretreated groups. Whereas pretreated CSF had a significantly lower protein concentration and lower rate of positive stained smear for bacteria (2).

Intracranial pressure [ICP] can be determined by obtaining an opening pressure at the time of the initial lumbar puncture (2). Clinical signs of elevated ICP should be monitored by an ICP monitoring device in order to prevent elevated intracranial pressure

which can result in life-threatening cerebral herniation (16, 17). The treatment of elevated intracranial pressure is outlined below (16).

1. Elevate the head of the bed 30°. Keep the head in the midline position.
2. Pentobarbital coma: Loading dose 5-10mg/kg IV. Therapy can be titrated to achieve a burst-suppression pattern on electroencephalogram [EEG]
3. Dexamethasome 0.15 mg/kg IV every 6 hours for the first four days.
4. Hyperventilation to maintain CO_2 partial pressure between 25-33mmHg.

Useful adjunctive therapy can be employed to reduce the amount of inflammatory cytokines and the number of leukocytes by administering dexamethasome [see figure 1 in appendix]. Dexamethasome use is indicated in infants and in adults with indications of raised intracranial pressure. It should be administered twenty minutes prior to the initiation of antibacterial therapy (17).

INTERNSHIP PRACTICUM JOURNAL

Wednesday, May 15, 2002

8:30-12:00 Attended Principle Investigator Training Session hosted by Medtrials.

Tuesday, May 28, 2002

8:00 a.m. Arrived at the UNTHSC Pediatrics and Allergy Clinics for first day of Internship Practicum. 8:45-9:00 Met with Sandra Powell. Shown around clinic and introduced to some of the clinic staff. 9:00-11:00 Contacted study participants by phone for follow-up questioning regarding the vaccination study they are enrolled in. 11:00-11:40 Prepared a CV to be added to the Regulatory Binder for inclusion on the 1572. 12:00-1:00 Lunch. 1:00-6:00 Worked on research proposal and journal entry.

Wednesday, May 29, 2002

8:00-9:00 Arrived at Pediatrics and Allergy Clinics and began to look up articles on med-line for bacterial meningitis. 9:00 Began patient follow-up calls on drug x trial to capture AE's and SAE's. 9:15-10:00 Visited drug x study subject with Lynette. Patients advised on a revised informed consent. Reviewed the patient diary for concomitant medications, possible adverse and serious adverse events and inspected the infant subject for rashes. In addition to the study procedure, Lynette gave patient's mother a table consisting of age appropriate dietary portions and food classes that can be tolerated by babies. Lynette administered the fourth month vaccination series. 10:00 Returned to place follow-up calls on the drug x study.

10:30-11:00 took a drug x study subject for a blood draw by the phlebotomist with Lynette. 11:00-12:30 Lunch. 12:30-1:30 Contact Robin Newman and Dr. Sheedlo about feedback on the proposal for the thesis. 1:30-3:00 Literature search.

Thursday, May 30, 2002

8:00-8:30 Looked over patient list with Sandra Powell for potential Formula study subjects. 8:30-11:00 Called drug x study subjects for follow-up questioning. Purpose of questioning is to capture adverse events [AE's] and significant adverse events [SAE's]. 11:00-12:30 Lunch. 12:30-1:30 Literature search. 1:30 Received call from TCOM admissions office notifying me of acceptance into the medical school.

Friday, May 31, 2002

8:00 Arrived at Pediatrics and Allergy Clinic. Checked my GroupWise e-mail. 8:30-11:00 Began subject follow-up calls for the drug x study. 11:00-11-30 Reviewed source documents with Lynette for errors and omissions. Focus of our review of the source documents was on recording AE's and SAE's, as well as, recording concomitant medications. The monitors on site flagged some of the discrepancies in the source documents compared to the data captured in the case report form [CRF]. Review of the other source documents was part of Lynette's standard review. 11:30-1:00 Lunch. 1:00-3:30 Continue placing follow-up calls for the drug x study to capture AE's and SAE's. 3:30-4:15 Met with Dr. Sheedlo concerning the thesis proposal and to get his thoughts and feedback on how to proceed.

Monday, June 3, 2002

8:30 Arrived at the Pediatrics and Allergy Clinic. 9:00-9:45 Began six-month follow-up calls for drug x trial. 10:00-11:00 Followed Dr. Fling on two allergy patient appointments. 11:00-11:30 Transferred data from six-month follow-up calls placed Friday into patient call logs. 11:30-1:30 Literature search. 1:30-2:15 Lunch. 2:30-4:00 Literature search.

Tuesday, June 4, 2002

8:30 Arrived at the Pediatrics and Allergy Clinic. Clinical Research Associate [CRA] is here to monitor the formula study. 9:00-12:30 Worked with Vicki Canon, the clinic's charge nurse in preparation for the Joint Commission on the Accreditation of Healthcare Organization [JCAHO]. We went through the drug closets assessing if medication and/or infant formula that ha reached their expiration date. After collecting the expired drugs and formula we recorded the drugs on a form. The form is delivered to the central pharmacy along with the drugs, to be recorded and destroyed by the pharmacy. 12:30-1:30 Lunch/ meeting with office staff to review JCAHO regulation questions that may be asked by JCAHO to the office staff. 1:30-2:30 Continued work on drug closets. 2:30-3:00 Accompanied Vicki Canon in an examination room. Vicki inserted a urinary catheter into a two-month-old female to collect a urine sample to be analyzed by the lab. 3:00-4:00 Went to attend an Institutional Review Board [IRB] meeting. The board chairperson was thirty minutes late. The layperson, required for all IRB full board meetings, was not present. The meeting had to be canceled, much to the disappointment of the reviewees. Thus the study proposals were not presented.

Wednesday, June 5, 2002

8:30-10:30 Arrived at Pediatrics and Allergy Clinic. Worked on literature search in office. 10:30-1:30 Walked over to Willis Library to pull journal articles and read for possible incorporation into thesis. 1:30-2:00 Lunch. 2:00-4:00 Sandra and Lynette were preparing for an investigator meeting in San Francisco. Continued literature search.

Thursday, June 6, 2002

9:00 Arrived at Pediatrics and Allergy Clinic to check freezer temperature and record in the temperature log. 9:15-1:30 Literature Review in the Willis library. 1:30-2:00 Lunch. 2:00-4:00 Literature review. 4:00 Checked and record freezer temperature in the freezer temperature log.

Friday, June 7, 2002

9:00 Arrived at Pediatrics and Allergy Clinic to check freezer temperature and record in the temperature log. 9:15-1:30 Literature review at the Willis library. 1:30-2:00 Lunch. 2:00-4:00 Literature review. 4:00 Checked and record freezer temperature in the freezer temperature log.

Saturday, June 08, 2002

10:00-11:30 Typed journal entries. 4:15-5:00 Typed journal entries.

Monday, June 10, 2002

8:30-9:00 Arrived at the Pediatrics and Allergy Clinic. Checked e-mail. 9:00-10:45 Worked on filling out six month subject visit forms and the accounting forms for final payment to subjects participating in the meningococcal vaccination clinical trial.

Made two copies of each, one copy went into the subject's source document, one to the site's file and the original to the Office for Clinical Trials on the UNTHSC campus. 10:45-11:30 Visited formula study subject with Sandra Powell for the four month check-up, blood draw and dispensing of final formula to the subject's mother. 11:30-1:00 Lunch. 1:00-4:00 Continued to work on the visit and payment forms.

Tuesday, June 11, 2002

8:45-11:00 Arrived at the Pediatrics and Allergy Clinic. Continued to work on the visit and patient payment forms. 11:00-1:00 Lunch. 1:00-3:30 Continued to work on the visit and payment forms.

Wednesday, June 12, 2002

8:00-9:00 Arrived at the Pediatrics and Allergy Clinic. Worked on journal and literature search. 9:00-12:40 Completed six-month case reports forms [CRF], AE/SAE and study termination sections for the meningococcal vaccination study. 12:40-2:00 Lunch. 2:00-4:00 Started literature review, received phone call friend had heart attack, work done for the day.

Thursday, June 13, 2002

8:30-12:00 Worked on completing CRFs and source documents for the meningococcal vaccination study. 12:00 Finished working for the day.

Friday, June 14, 2002

8:15-9:30 Arrived at the Pediatrics and Allergy Clinic. Checked e-mail and performed literature search. 9:30-10:00 Searched for a current phone number on the internet for a subject that we have not been able to reach for the six-month follow-up.

I was able to find a phone number and left a message. Sandra Powell had previously sent a letter via certified mail. The subject's wife received the certified letter, however the subjects has not contacted Sandra Powell to complete the follow-up. Completed a few more source documents and CRFs for patients that have been recently contacted for their six-month follow-up. 10:00-11:30 Reviewed the meningococcal vaccination study protocol. Went to Dr. Fling to inquire about some of the technical aspects of the study. 11:30-12:30 Lunch; end of day; visit friend in hospital.

Saturday, June 15, 2002

Entered the week's journal entries into Word document.

Tuesday, June 18, 2002

8:15 Arrived at the Pediatrics and Allergy Clinic. Checked e-mail and looked for literature on Center for Disease Control and Prevention website for meningococcal meningitis epidemiology information. 9:00-12:00 Monitor for the vaccination study arrived for two-day visit to conduct study closeout. There was not a lot for me to do at the clinic, as the monitor occupies the office where I usually perform assigned tasks. Went to library to look for more journal articles.

Wednesday, June 19, 2002

8:15 Arrived at the Pediatrics and Allergy Clinic. Began working on source documents and CRFs flagged by the Allison, the CRA monitoring the meningitis vaccination trial. Most frequently flagged items were improper reporting of AE's and SAE's in the CRFs, AE's and SAE's not properly documented in the source documents and missing signature of the Principle Investigator [P.I.]. 11:30-1:00 Manually typed three SAE reports.

Thursday, June 20, 2002

9:00-11:00 Arrived at the Pediatrics and Allergy Clinic. Completed an online tutorial required for all persons working in clinical trials at the UNTHSC. The tutorial involved issues of subject safety, the elements of informed consent and the Belmont Report's Respect for Persons, Beneficence and Justice in performing clinical research on human subjects. The documentation of completion is then submitted along with a CV to the Office of Clinical trials on the campus of the UNTHSC.

Friday, June 21, 2002

9:00-9:30 Arrived at Pediatrics and Allergy Clinic. Dr. Fling, Sandra Powell and Lynette Lode are away at an investigator's conference in Virginia. Dr. Al Levine unlocked the investigative drug room to allow me to record the freezer temperature log. Also, I checked to see if a shipment of investigational infant formula had arrived. 9:30-10:30 Worked on journal entries. 10:30-12:00 Literature review. 12:00-1:00 Lunch. 1:00-2:30 Literature review. 2:30 I returned to internship site to record the investigational drug freezer temperature.

Monday, June 24, 2002

9:00-10:00 Arrived at the Pediatrics and Allergy Clinic. Sandra Powell and Lynette Lode are away at the investigator's conference. Recorded the temperature of the investigational drug freezer in the temperature log. Unpacked a shipment of investigational infant formula and placed the cases in the investigational drug cabinet.

Faxed confirmation of receipt of the investigational product to the sponsor to verify product remained within the specified temperature parameters and that the quantity of product shipped is as stated on the manifest matched the quantity of product received. 11:00-1:00 Returned home to work on practicum. 1:00-1:30 Lunch. 1:30-4:30 Continued work on practicum.

Tuesday, June 25, 2002

8:15-11:00 Arrived at the Pediatrics and Allergy Clinic. Online Literature search using Ovid to search for relevant journal articles. 11:00-12:00 Lunch. 12:00-2:30 Literature review of journals found earlier in the morning. 2:30-3:30 Returned home. 3:30-5:00 Worked on practicum.

Wednesday, June 26, 2002

8:45-12:30 Arrived at Pediatrics and Allergy Clinic. Worked on contacting the few remaining subjects on the meningitis vaccination study who have not completed the six-month follow-ups. I searched for current phone numbers, mailing addresses and e-mail addresses on the internet. Found a couple of phone numbers and left messages to contact Sandra Powell to answer a few follow-up questions. Completed follow-up on one subject, e-mailed a few others. 12:30-1:30 Lunch. 1:30-3:00 Worked on practicum.

Thursday, June 27, 2002

8:30-11:30 Arrived at Pediatrics and Allergy Clinic. I Pulled medical charts of several patients to screen for match to inclusion/exclusion criteria for investigational formula trial. Unpacked shipping boxes supplied by the sponsor.

I rearranged the shelving and the rest of the investigational drug room to make space for the additional shipping boxes. 11:30-12:30 Lunch. 12:30-5:00 Worked on writing practicum.

Friday, June 28, 2002

8:15-9:30 Arrived at the Pediatrics and Allergy Clinic. Sandra Powell did not come in today. 9:30-11:00 Literature search in the Willis Library. 11:00-12:00 Lunch. 12:00-4:00 Worked on writing practicum.

Monday, July 01, 2002

8:30-10:40 Arrived at the Pediatrics and Allergy Clinic. I updated the site's Trials Ongoing board. The board summarizes the trials ongoing by the protocol number, trial name and code, the sponsor, the enrollment number, the number enrolled, the number screened, the Clinical Research Organization [CRO], and the investigator. 10:40-12:00 Walked over to the Willis Library to make copies of journals found on an Ovid search. 12:00-1:30 Returned home and have lunch. 1:30-3:30 Worked on typing practicum. 3:30-5:30 Typed journal entries from previous week to today.

Tuesday, July 2, 2002

8:30-10:00 Arrived at the Pediatrics and Allergy Clinic. I attempted to contact the remaining subjects for the six-month follow-up questionnaire to capture AEs and SAEs. Was able to contact and obtain the required information for one subject. Spoke to a relative of another subject who said they would try to get subject to call in to the site and complete the study. 12:00-1:00 Lunch. 1:00-1:30 Spoke with TCOM financial aid office. 1:30-4:30 Worked on typing practicum. 5:30-8:20 Continued writing practicum.

Wednesday, July 03, 2002

9:00-2:00 Arrived at the Pediatrics and Allergy Clinic. Today is the last day at the internship site. Sandra Powell asked me if I would be interested in working some part-time hours [5-10 hours per week] by helping with future studies. I told her that I would be interested if my school schedule allowed for it. Began working on copying the signed informed consents forms and non-coercion forms, when applicable, from the subjects files. The copies are to be added to the regulatory binder. I will return Monday, July 8, for a final meeting with Dr. Fling, Sandra Powell and Lynette Lode.

DISCUSSION

A brief overview of the different mechanisms involved in the immune response against polysaccharide vaccine versus those seen in the conjugate polysaccharide will first be discussed. Then I would like to review the activities involved at the internship site along with a review of the seminar hosted by MedTrials on the topic of the protection of human subjects in investigational product research.

Immune Response

Conjugated meningococcal vaccines work by covalently linking the outer membrane polysaccharide to an immunogenic protein such as diptheria toxoid (9). These new conjugated vaccines are considered superior to polysaccharide vaccines by being able to induce a T-dependent immunologic response and create immunological memory. The polysaccharide vaccine has demonstrated an induction of a T-independent immunologic response with a shorter duration of immunological effectiveness.

The outer membrane capsular polysaccharide is highly repetitive structure that has no intrinsic B-cell-stimulating activity and is termed a TI-2 antigen (9). B-cells are immune cells that specialize in antibody production. The TI-2 antigen can activate only mature B cells; immature B cells are inactivated by such repetitive epitopes. This might be why infants do not make antibodies to polysaccharide antigens efficiently; most of their B cells are immature. Responses to several TI-2 antigens are prominent among B-1 cells, which comprise an autonomously replicating subpopulation of B cells, and among marginal zone B cells, another unique subset of non-recirculating B cells that line the

border of the splenic white pulp. Although B-1 cells arise early in development, young children do not make a fully effective response to carbohydrate antigens until about five years of age. On the other hand, marginal zone B cells are rare at birth and accumulate with age; they may thus be responsible for most physiological TI-2 responses, which also increase with age. These B-cells produce both IgM and IgG antibodies. It is the IgG antibodies are most effective at ridding the body of encapsulated bacteria by opsonized inducement of phagocytosis by macrophages and neutrophils and the recruitment of late complement pathway formation of membrane attack complexes. IgM is effective in neutralizing bacteria by forming an opsonizing coat of antibody around the organism, preventing it from interacting with host cell surface receptors (9). The phenomenon of hyporesponsiveness to subsequent polysaccharide vaccination is still unclear. What is clear is that immunological memory is not created.

The process of clonal expansion and creation of immunological memory must proceed through the response of immature B-cells to a T-cell (9). The process of immature B-cell recognition of antigen does not occur with TI-2 class antigens such as meningococcal capsular polysaccharides. T-cell recognition of the major hisocompatability complex [MHC II] that displays the antigenic particle on the surface of the B-cell are required for stimulating clonal expansion and subsequent selection of memory B-cell production.

B-cell responses to TI-2 antigens provide a prompt and specific response to an important class of pathogen (9). Bacterial pathogens surrounded by a polysaccharide capsule enable them to resist ingestion by phagocytes.

The bacteria not only escape direct destruction by phagocytes but also avoid stimulating T-cell responses through the presentation of bacterial peptides by macrophages. Antibody that is produced rapidly by B-1 cells in response to this polysaccharide containing capsule without the help of peptide-specific T cells can coat these bacteria, promoting their ingestion and destruction by phagocytes through opsonization by antibody and/ or opsonization by antibody/ complement complex (9).

The importance of the complement pathway in infection by *N. meningitidis* is noted in a large population study done in Japan, concerning late complement deficiencies, where endemic *N. meningitidis* infection is rare (9). The results of the study showed that the risk each year to a normal person of infection with this organism is approximately 1/ 2,000,000. This compares with a risk of 1/ 200 in the same population to a person with inherited late complement deficiency of the membrane attack complex. This indicates that host defense against these bacteria, which are capable of intracellular survival, is mediated by extracellular lysis by the membrane-attack complex of complement (9).

Conjugate vaccines are able to create immunologic memory by a unique method of stimulating a T-dependent response (9). The B-cell recognizes the antigen, in this case the meningococcal capsular polysaccharide diptheria toxoid conjugate. The B-cell membrane bound antibody-antigen complex is internalized by the cell and degraded. The degradation components, specifically of interest in this case, the diptheria toxoid, are then added to the major histocompatability complex [MHC II] and displayed on the surface of the B-cell.

Helper T-cells generated by an earlier vaccination against diptheria toxoid recognize the complex on the B-cell surface and activate the B-cell to produce polysaccharide antibodies, undergo proliferation, and become plasma cells or specialized memory B-cells (9).

Currently, mono- and bivalent meningococcal conjugated vaccines are being marketed. The British government implemented a comprehensive meningococcal conjugate vaccination program using a mono-valent serogroup C vaccine, as the majority of the meningococcal cases in Britain are due to the serogroup C strain of *N. meningitidis*.

There are no vaccines against the serogroup B strain of meningococcus that are thought to be safe and effective (15). MenB [meningococcus serogroup B] polysaccharide is poorly immunogenic in humans. Also, there is a fear that using this polysaccharide as a vaccine would hide risks of immunological tolerance because the homopolymer of α [2 →8] N-acetyl-neuraminic acid might cross-react with polysialic acids of embryonic neural cell adhesion molecules. Perhaps an autoimmune process would be triggered, and vaccine-induced antibodies might interfere with the functions of the polysialylated protein components of the brain. The relevance of the theory has been questioned, but since it is very difficult to prove or disprove, it has blocked much of the research on MenB polysaccharide. Nevertheless, induced anti-MenB polysaccharide antibodies are bactericidal in the presence of human complement (15). A serogroup B sub-type specific meningococcal outer membrane protein vaccine has been developed and studied in Cuban infants, adolescents and adults (13). This effort resulted in the discovery of the development of an immunological response and bactericidal activity specific to the

serogroup B vaccine sub-type but with limited serogroup B other sub-type cross reactivity (13). Future study is planned to look for a more globally responsive serogroup B vaccine.

Internship Activities

The site of the internship was at the Pediatrics and Allergy Clinic in the Patient Care Center on the campus of the University of North Texas Health Science Center. The internship was under the supervision of Dr. John Fling, Sandra Powell, Certified Clinical Research Coordinator [CCRC] and Lynette Lode [CRC].

Activities of the internship were focused on the clinical trial of a tetravalent meningococcal [A, C, Y and W-135] diptheria conjugate vaccine's safety, immunogenicity and lot consistency among healthy adults compared to a tetravalent meningococcal [A, C, Y and W-135] polysaccharide vaccine currently marketed.

Most of my efforts involved completing six-month follow-up contacts by phone. The six-month follow-up is performed to capture adverse events [AE's] and significant adverse events [SAE's]. The principal investigator, Dr. John Fling, then determines whether an event[s], AE or SAE, captured through the designed questions is likely related to the investigational drug. After completing the six-month follow-ups, I worked on transcribing the six-month follow-up information, now a part of the source document, into the case report form [CRF]. Soon thereafter the clinical research associate [CRA] from the clinical research organization [CRO], hired by the sponsor to monitor its investigational drug trial, arrived to monitor the trial. Several errors were discovered in the transcription of the source document to the CRF. Most of the errors involved my

transcribing of adverse events into the CRF that were not determined by the PI to be related to the drug.

In addition to data collection and transcription, I participated in several other research-related activities. These activities included reviewing the patient appointment schedule for age related inclusion/ exclusion criteria for the investigational infant formula study enrollment, accompanying Ms. Lode to issue a revised copy of an informed consent to the mothers of patients enrolled in the infant formula study, shadowing Ms. Lode on the follow-up visits of the childhood vaccination study, completing subject compensation forms for the meningococcal vaccination study, and making copies of the signed final version of the vaccination study's informed consent. The informed consents are then added to the regulatory binder per the sponsor and/or the CRO. Along with the informed consent the signed statement of non-coercion was added to the CRF when applicable. The non-coercion statement is a declaration that a student or employee is freely volunteering to take part in the study and is not being pressured in any manner to take part in the study. In addition, I monitored and recorded the investigational drug freezer temperature twice daily while Dr. Fling, Ms. Powell and Ms. Lode were away at an investigators conference. I also received supplies sent by the sponsor, verified the shipment's manifest and that the proper temperature was maintained for temperature sensitive items. A form was then completed and faxed to the sponsor verifying the correctness of the shipment.

Prior to the internship I was invited to attend a training session with Dr. Fling. The three and a half-hour session was hosted by MedTrials, a clinical research organization [CRO] that offers graduate level courses and seminars on the conduction of clinical drug and device trials in addition to their traditional CRO activities. The subject was, *The Protection of Human Subjects*, covering Good Clinical Practice Applications and Ethics, Research and the Law. Good Clinical Practice Applications were outlined according to Title 21 of the Code of Federal Regulations which governs how the development and investigation of drugs and devices are to be carried out while protecting the "rights of the subjects to privacy, autonomy, beneficence, safety and to protect the integrity of clinical data" (11). Investigator accountability was also emphasized, as was drug accountability.

Ethics in clinical research was covered in order to define good science in the design of a study. Four areas were defined for maximizing the overall "assuring of substantial evidence of effectiveness as called for in the U.S. Federal Food, Drug, and Cosmetic Act of 1938 as amended, by providing '...evidence consisting on adequate and well-controlled investigation, including clinical investigation, by qualified scientific experts, that proves the drug will have the effect claimed by its labeling'"(11, 33). The four areas are as follows (33).

1. Well-designed trials
2. Selection of qualified investigators
3. Thorough study initiations
4. Closely monitored, well-controlled trials.

The Belmont Report (14) was cited for the manner in which it defines three basic principles, among those generally accepted in our cultural tradition that are particularly relevant to the ethics of research involving human subjects: respect for persons [protection of a research subject's autonomy], beneficence [do no harm and maximize benefits and minimize risk], and justice [the portion of society participating in the study should also be able to benefit from the product if it were to be marketed and one segment of society should not be unduly burdened in the investigation of the product, that is, those of lower socioeconomic status].

The principles and elements of informed consent under 21 CRF section 50, was also a major topic of discussion (33). Informed consent should be thought of as an ongoing process throughout the participation. When designing a consent form one must: keep the language simple, with a maximum reading level not to exceed eighth grade, not include exculpatory language, avoid foreign terms or phrases, avoid jargon, define all terms listed, avoid colloquialisms, and when possible use a qualified translator if English is second language. There are eight elements with an additional six that must be part of all informed consents drafted by the principal investigator and approved by the institutional review board [IRB]. The elements are as follows (33).

1. A statement that the study involves research, an explanation of the purpose of the study, expected duration of the patient's participation, subject's responsibilities, an explanation of procedures to be followed and identification of experimental procedures.
2. A description of any reasonably foreseeable risks or discomforts.

3. A description of any reasonably foreseeable benefits from the research to the subject.
4. A disclosure of any alternative procedures of possible benefit to the subject.
5. A description of the extent of confidentiality and that the FDA may inspect the records.
6. For research involving more than minimal risk, an explanation as to whether any compensation and any medical treatments are available should injury occur and, if so, what they comprise or where further information may be obtained.
7. A contact person for answers to pertinent questions about the research and research-subject's rights and whom to contact in the event of an injury to the subject.
8. A statement that participation is voluntary, that refusal to participate will involve no penalty or loss of benefits to which the subject is otherwise entitled and that the subject may discontinue participation at any time without penalty or loss of benefits to which he is otherwise entitled.

Additional elements of informed consent are as follows (33):

1. A statement that the particular treatment or procedure may involve unforeseeable risks.
2. Anticipated circumstances under which the subject's participation may be terminated by the investigator without regard to the subject's consent.
3. Any additional cost to the subject that may result from participation in the research.

4. The consequences of a subject's decision to withdraw from the research and procedures for orderly termination of participation by the subject.
5. A statement that significant new findings developed during the course of the research which may relate to the subject's willingness to continue participation will be provided to the subject.
6. The approximate number of subjects involved in the study.

Financial disclosure of the investigator and sponsor was discussed in how it relates to possibly creating investigator bias. The FDA requires that significant financial holdings owned by the investigator or investigator's family in addition to significant payments of other sorts made to the investigator by the sponsor, proprietary interest in the test product, and the steps taken to minimize the potential for bias (33). FDA Form 3455 is to be completed disclosing financial interests and the steps taken to minimize bias.

Individuals and corporations attempting to bring drugs and devices to the marketplace invest huge sums of money. In the case of a new drug, it generally takes eight to ten years from the initial synthesis of the test product to approval by the FDA. The amount of time and capital involved in the development and testing of an investigational product makes it imperative that the product will be safe, effective and approvable by the FDA.

What I take away most from this internship and Masters program is the emphasis on research subject safety, privacy and guarding the integrity of the research data. The Title 21 Code of Federal Regulations has been developed to safeguard the integrity of medical research and moreover the safety and individuals rights of the subjects involved in the research.

The short seminar on, The Protection of Human Subjects, was a great refresher of the material covered in the graduate course offered by MedTrials and UNTHSC. It is my opinion that all investigators wishing to conduct research on human subjects should be required to complete training over similar material covered in the graduate course, Introduction to Clinical Research and Studies, taught by the staff at MedTrials.

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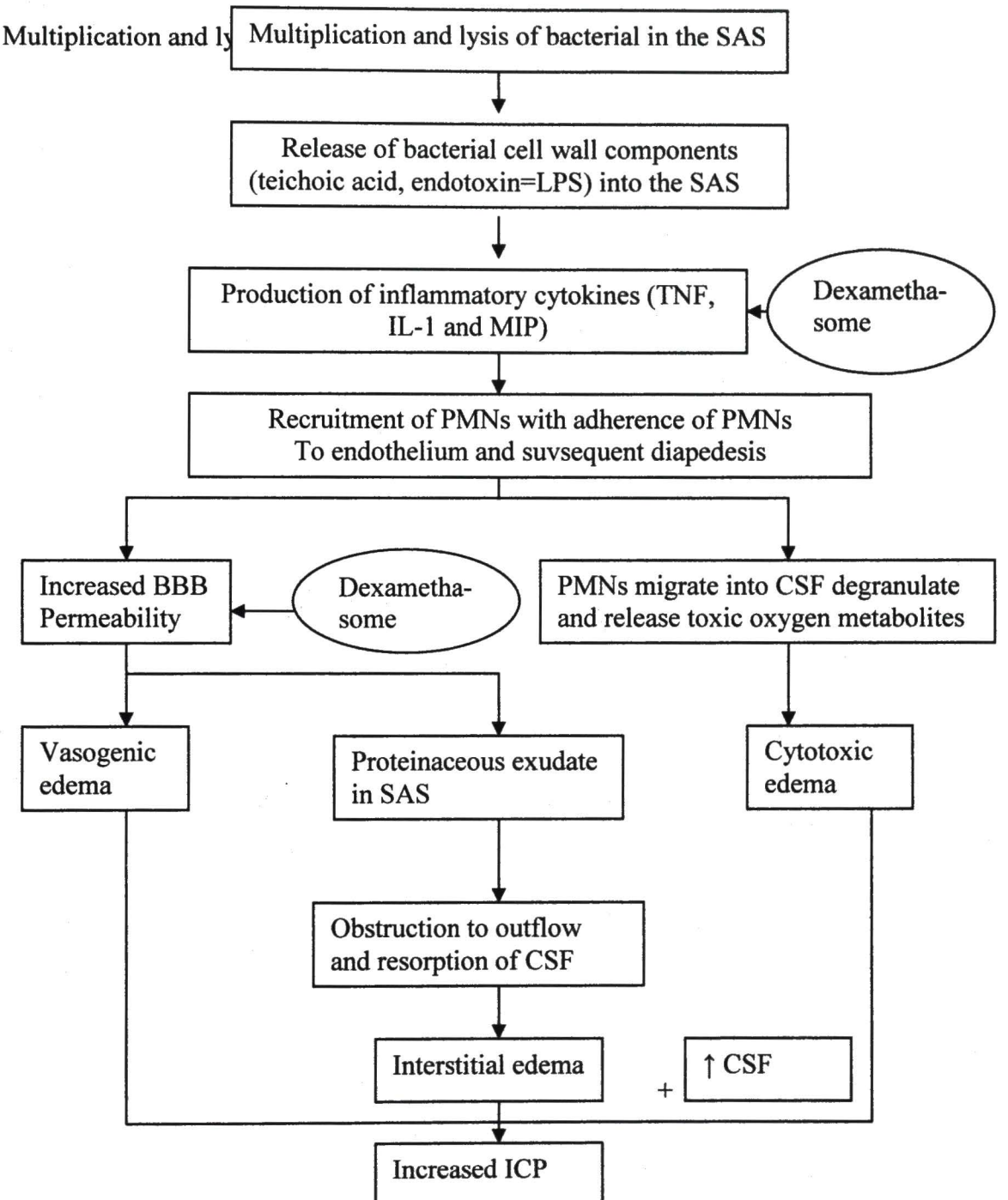
APPENDIX

Figure 1. Dexamethasome in adjunctive therapy

Pathophysiology of Bacterial Meningitis



SAS= Subarachnoid Space, LPS= Lipopolysaccharide, TNF= Tumor Necrosis Factor α, IL-1= Interleukin 1, MIP= Macrophage Inflammatory Protein, ICP= Intracranial Pressure (2)