Journal of Pediatric Sciences

The Use of Penicillin for the Prevention of Pneumococcal Infection in Pediatric Patients with Sickle Cell Disease

Bola Lawuyi, Jamie Basham, Tsz-Yin So

Journal of Pediatric Sciences 2015;7:e246

How to cite this article:

Bola Lawuyi, Jamie Basham, Tsz-Yin So. The Use of Penicillin for the Prevention of Pneumococcal Infection in Pediatric Patients with Sickle Cell Disease. Journal of Pediatric Sciences. 2015;7:e246

REVIEW ARTICLE

The Use of Penicillin for the Prevention of Pneumococcal Infection in Pediatric Patients with Sickle Cell Disease

[1]Bola Lawuyi, [2]Jamie Basham, [3]Tsz-Yin So

[1]Cardiology Specialty Resident, Department of Pharmacy, The Ohio State University Medical Center, USA
[2]Pharmacy Resident, Department of Pharmacy, Saint Thomas West Hospital, USA
[3]Pediatric Clinical Pharmacist, Department of Pharmacy, Moses H. Cone Hospital, USA

Abstract:
Background: *Children with sickle cell disease and asplenia are at a 30-600-fold increased risk of acquiring Streptococcus pneumoniae infection when compared to children without this disease. Prevention of such infection is vital in this population.* ***Data sources:*** *We searched PubMed with the key words penicillin, pediatric, pneumococcal vaccines, and sickle cell anemia and selected publications that were deemed appropriate.* ***Results:*** *Penicillin prophylaxis has been shown to be efficacious in children less than 5 years of age, but its therapeutic benefits in children over the age of 5 remain unknown. One Cochrane review and three randomized trials evaluated the efficacy of penicillin prophylaxis across different age groups in patients with HbSS and Sβ0Thal sickle cell disease. The rate of pneumococcal infection in the three trials combined showed a reduction (OR 0.37, 95%CI 0.16-0.86) of Streptococcus pneumoniae infection in children less than 5 years old who had received penicillin prophylaxis. The risk of such infection was lower in children over the age of 5. Long-term prophylaxis of SCD patients with penicillin poses a concern of drug-resistant strain of the S. pneumoniae and this risk continues to be a significant area of controversy.* ***Conclusions:*** *Infection caused by Streptococcus pneumoniae is reduced with the use of penicillin prophylaxis under the age of 5. It may be safe to discontinue prophylaxis in patients without splenectomy and history of severe pneumococcal sepsis once the patient has reached 5 years old due to unclear benefit and the potential risk of developing penicillin induced-streptococcal resistance.*

Keywords: *Penicillins, pediatric, pneumococcal vaccines, sickle cell anemia*
Corresponding author: *Tsz-Yin So, PharmD, BCPS, Pediatric Clinical Pharmacist, Department of Pharmacy, Moses H. Cone Hospital, 1200 N. Elm Street, Greensboro, NC 27401-1020, Telephone# 336/832-6166, Fax 336/832-7198, E-mail tszsounc@gmail.com*

Introduction

Sickle cell disease (SCD) is the most common inherited blood disorder in the United States (US); it affects about 70,000 to 80,000 Americans. The disease is estimated to occur in 1 in every 500 African Americans and 1 in every 1,000 to 1,400 Hispanic Americans [1]. Sickle cell disease is a genetic hemoglobin disorder. The sickle cell gene carries a single mutation leading to the substitution of valine for glutamic acid at the sixth amino acid position of the β globin chain. When the hemoglobin molecule is in the deoxygenated state or when the oxygen saturation is lowered, this change in the amino acid leads to polymerization of the hemoglobin resulting in deformity. These sickled cells become trapped in the capillaries and adhere to blood vessel linings preventing blood flow to various vital organs [2].

The inheritance of various sickle cell genes results in

differing variations of sickle cell disease. The sickle cell trait is the result of a single sickle cell gene. Children with sickle cell trait are usually without symptoms of the disease because they have both destructive and normal hemoglobin. Sickle cell anemia occurs when the child has inherited the sickle cell gene from both parents. Most of the normal hemoglobin is replaced with the sickle hemoglobin (HbS). This is referred as HbSS. It is the most common and most severe form of the sickle cell variations.

Patients with sickle cell hemoglobin SC disease have one copy of both HbS and HbC genes, which is referred as HbSC. Children with sickle cell HbSC disease may suffer from a milder form of some of the complications that are usually experienced by patients with sickle cell HbSS disease. Individuals with Sβ0Thal have one HbS gene and one gene of hemoglobin beta-thalassemia. The beta-thalassemia gene (β0Thal) indicates that the patient lacks normal hemoglobin A [3].

Sickle cell disease was once considered a childhood illness since relatively few patients survived very far into adulthood. Although improved over the past few years, the life expectancy only averages 42 and 48 years of age in men and women, respectively [1]. Hallmark complications include vaso-occlusive crises leading to organ damage and severe pain. Pulmonary hypertension, stroke, anemia, kidney problems, liver problems, gallbladder disease, and spleen damage are also common complications [4]. The immune system of patients with splenic dysfunction has reduced ability to clear antigens. SCD patients are therefore most susceptible to respiratory infections and septicemia, especially those that are caused by encapsulated bacteria such as pneumococci [5]. The most common cause of death in children with sickle cell disease is infection, and it is the most apparent in patients with genotype SS and Sβ0Thal [6].

The U.S Preventive Services Task Force recommends early screening in childhood for sickle cell disease. Screening has become widely available due to the positive evidence that early detection of sickle cell disease followed by prophylactic oral penicillin can reduce the risk of deadly infections in this population [7]. In addition, educating parents about the early warning signs of infections may hasten appropriate care. The American Academy of Pediatrics supports these suggestions and recommends the use of penicillin prophylaxis in children with sickle cell disease under the age of five and in older children who have had a previous severe pneumococcal infection or have functional/surgical asplenia [8]. Although the efficacy of penicillin prophylaxis has been well established for pneumococcal infection in sickle cell patients, whether continuing prophylaxis beyond 5 years of age and the influence of penicillin on inducing streptococcal resistance remain to be areas of controversy [9]. This article will discuss the pathophysiology, pathogenesis, virulence factors, epidemiology, prevention and resistance of pneumococcal disease in patients with SCD and the efficacy of penicillin prophylaxis for this potentially fatal infection.

Immune function of the spleen

Organ damage in sickle cell disease results when sickled red blood cells block blood vessels, reducing oxygen-rich blood flow. The most common and earliest organ to be affected in sickle cell anemia is the spleen. The progression to asplenia begins early in life when the spleen becomes enlarged due to entrapped red blood cells. It then undergoes atrophy leading to a shrunken and nonfunctional spleen. In other cases, the enlarged spleen persists, requiring splenectomy to prevent complications such as splenic abscesses and severe anemia. Studies indicated that by 5 years of age, 94% of patients are affected with the absence of splenic function [10].

Spleen acts as a major filter of blood in the body; it filters about 5% of the total cardiac output every minute. During this process, if the blood contents are discharged into the cords of the red pulp, these blood cells contents, which potentially include bacteria, can be in prolonged and close contact with the macrophages in the splenic cells. This interaction will in turn activates the phagocytosis of the pathogens. On the other hand, the white pulp, which is divided into T lymphocyte zones and follicles that contain B cells, macrophages and dendritic cells, helps clear the lymphocytes from the blood so that the spleen can start its own adaptive immune response. By producing anti-polysaccharide immunoglobulin M (IgM), the B cells and the dendritic cells can capture antigens, especially the poorly opsonized encapsulated bacteria such as Streptococcus pneumoniae (S. pneumoniae), Haemophilus influenzae type b (Hib) and Nesseiria meningitidis. These cells can also promote both T cell-dependent and –independent immune response. Thus, a defective spleen, like in the case of patients with sickle cell disease, can lead to increased risk of bacterial infections and autoimmunity [11].

Pathogenesis, pathophysiology and virulence factors of pneumococcal infection

S. pneumoniae is an organism that is colonized in the upper respiratory tract of many healthy individuals. The attachment of such pathogens to the respiratory tract is thought to be mediated by a disaccharide receptor on fibronectin of pharyngeal epithelial cells [12]. The mechanisms by which these pathogens translocate from the nasopharynx to the lung or the blood are not well understood. The hosts' immune and clinical status and the virulence of the strains of the pathogens play an important role in determining whether or not they will be invasive or just be confined to the nasopharynx. For example, the reduction in mucosal secretion, ciliary transport, and the amount of secretory immunoglobulin A (IgA) in the respiratory tract may increase a host's risk of having the pneumococci gain access to the bronchi and the lungs [13].

Unrestricted and potentially harmful proliferation of pneumococci at the site of infection does not usually occur in healthy individuals, but this may not be the case in SCD patients. Several theories have been postulated by scientists explaining the immune response to pneumococci in patients with SCD. A defect in the alternative pathway of complement activation has been observed, but the exact site of this defect is not well known. Patients with SCD possess reduced opsonic activity against S. pneumoniae [14]. This reduced activity is more pronounced in patients < 2 years of age. Chudwin et al [15] found that the type 7 pneumococcal polysaccharide antibody concentration and opsonic activity of 6 patients less than 2 years of age did not differ significantly after immunization, but this was not the case in patients >/= 2 years old. Also, the activated complement 3 (C3) generated by the alternative pathway of complement activation in SCD patients is thought to be ineffective in helping with the phagocyctosis and the intracellular destruction of the pneumococci by poly-morphonuclear leukocytes [16]. Even though the sera from SCD patients facilitates adherence to the pneumococci, it does not increase uptake and killing of the pathogens. Bjornson et al [17] found that adherence was observed to be in the low-normal range in SCD patients in the absence of added immunoglobulin. The anaerobic intracellular and extracellular conditions in SCD patients are also favorable factors in increasing the risk of pneumococcal infection. These conditions can occur in areas of hypoxia (e.g. spleen, lung and bone) due to the reduction in hemoglobin S saturation [18]. Lastly, researchers have postulated a causative role for other immune deficiencies causing the increased susceptibility of SCD patients to acquire pneumococcal infection. For example, Dimitrov et al [19] observed that the function of leukocytes in SCD patients can be defective, and there can be an inadequate amount of intracellular hydrogen peroxide and hexose monophosphate shunt activity during phagocyctosis.

Infections with encapsulated bacteria such as pneumococcal species is thought to be the most detrimental in patients under the age of two. The capsule allows for avoidance of phagocyte killing. In healthy adults, however, this can be overcome by antibody production. Children less than 3 years of age are especially prone to these infections due to the fact that their immune system has reduced ability to target encapsulated pathogens [20]. Infants, for example, physiologically lack IgM memory B cells, and therefore, cannot produce natural antibodies to combat the encapsulated pathogens mentioned previously. Injury to the spleen from sickle cell disease further impairs the development of these IgM memory B cells and increases the risk of infection [11].

Other major virulence factors of S. pneumoniae include, and are not limited to, its complement factor H-binding component, autolysin, and peptide permeases. The complement factor H-binding component may inhibit the hosts' complement activation and phagocyctosis of the pathogens. On the other hand, autolysin can promote the release of pneumolysin, which, at low concentration, can inhibit bactericidal activity of polymorphonuclear leukocytes. It can also inhibit ciliary movement of epithelium preventing the clearance of the pathogens from the respiratory tract. Besides this, peptide permeases can enhance adhesion of pneumococci to endothelial cells. All these factors play an important role in the virulence of these common but potentially deadly bacteria in a human host [21].

Epidemiology of pneumococcal infection in sickle cell disease

S. pneumoniae is the cause of infection in more than 70% of all cases in children with sickle cell disease [22]. The chance of acquiring pneumococcal meningitis in children with sickle cell disease is 36 times greater than in African American children not diagnosed with sickle cell disease and 314 times greater than in Caucasian children [22]. The average incidence of meningitis in patients with sickle cell

anemia is about 30 cases per 100,000 population and is only 1.36 cases per 100,000 population in healthy subjects [22]. The relative risk of infection caused by S. pneumoniae in young patients with sickle cell disease compared to healthy children is about 300, with a mortality rate of 15% [11].

Pneumococcal septicemia progresses rather quickly, often escalating from onset to death in less than 12 hours [5]. The most prevalent pneumococcal serotypes affecting children with sickle cell disease are types 6, 14, 18, 19, and 23. These are also the serotypes that clinical trials typically report in vaccine failures [22].Thirty-five percent of pneumococcal sepsis and 10% of pneumococcal meningitis resulted in fatality before penicillin prophylaxis becoming a routine practice [23]. During these early years, infection among children younger than 5 years old occurred at 3.2 to 6.9 events per 100 patient years [23]. At this same time, children from 6 to 19 years old suffered from 0.1 to 0.2 events per 100 patient years [23]. This shows the need for prophylaxis in our younger population. The risk of severe S. pneumoniae infection is thought to decrease with age; however, those aged 5 to 21 years are still at an increased risk of 10-40 times that of the general population in obtaining an infection. One potential reason that the risk of pneumococcal infection is reduced in older patients is probably due to the completion of the pneumococcal vaccine series. Also, as mentioned earlier, patients less than 2 years old may potentially have more pronounced reduction in opsonic activity. In fact, Chudwin et al [15] found that the type 7 pneumococcal polysaccharide antibody concentration and opsonic activity of 6 patients less than 2 years of age did not differ significantly after immunization, but this was not the case in patients >/= 2 years old. Because of this, it has been postulated that penicillin prophylaxis could be discontinued in early childhood [6,9].

Pneumococcal colonization in children with sickle cell disease

The risk of developing invasive infection caused by S. pneumoniae is greatly associated with nasopharyngeal colonization [24,25]. The prevalence of colonization in the general pediatric population is believed to be similar to that of the sickle cell population [26,27]. According to Battersby et al [28], nasopharyngeal colonization with S. pneumoniae is believed to be around 30 – 40% in children with SCD. However, in one study by Fonseca et al [24], the prevalence of such colonization was found to be only 13.3% in Sao Paulo, Brazil, whereas the rate of colonization was found to be around 27% at an academic center in San Antonio, TX, United States [29].

The rate of nasopharyngeal colonization with S. pneumoniae is influenced by different factors ranging from geographic location to study population. In one study, patients less than 2 years of age ($p < 0.001$) and patients attending day care for more than 20 hours per week ($p = 0.00005$) were found to have increased incidence of pneumococcal colonization [30]. Fonseca et al [24] also found that patient aged </= 2 years old were at risk for colonization when compared to other age groups. This study did not show day-care attendance as a risk factor probably due to the low percentage of patients who attended such environment in the study. Regardless of the difference in findings, preventive measures should be implemented especially for these groups of patients with SCD.

The effect of pneumococcal vaccination on the nasopharyngeal carriage of S. pneumoniae and other respiratory pathogens is of great interest. Some reports demonstrated that even though pneumococcal vaccine helps reduce the carriage of vaccine-type S. pneumoniae, it potentially may increase the carriage of other pathogens such as H. influenzae [31]. Researchers like van Gils et al [32] also observed that there was a temporary increase in Staphylococcus aureus (S. aureus) colonization at 12 months of age in infants who had finished 3 doses of the 7-valent pneumococcal conjugate vaccine. On the contrary, pneumococcal vaccination was not shown to have any effect on other pathogens' carriage rates (specifically H. influenzae, Moraxella catarrhalis, and S. aureus) in Fijian infants [33]. Further researches are needed to determine if this effect of pneumococcal vaccine on the carriage rates of other pathogens truly exists.

Prevention of pneumococcal infection in sickle cell disease

In order for prophylactic agents to be initiated, a diagnosis of SCD has to be made. Newborn screening programs for hemoglobinopathies broke ground in the 1970s. Health departments implemented screening programs in New York in 1975, Illinois in 1989, and California in 1990 [34]. Other states soon followed suit. The findings of these screening reports indicated low mortality rates when children were diagnosed with SCD soon after birth [34]. Newborn screening is very useful and allows physicians to initiate infection prevention with penicillin and pneumococcal vaccination at age appropriate times. Another option

is prenatal screening when the mother can find out whether she has HbS or other hemoglobinopathies [35]. If she has abnormal hemoglobin, the newborn will then be tested. If her hemoglobin is normal, the newborn will not be tested. Either way, screening in some shape or form is important to identify those children with sickle cell disease before 3 months of age when their symptoms may start to surface.The mainstay of evidence for penicillin prophylaxis comes from the Prophylactic Penicillin Study (PROPS) trial, which concluded that penicillin V potassium 125 mg orally twice daily (BID) is recommended in those patients less than 3 years of age [5]. Patients aged 3 to 5 should receive penicillin V potassium 250 mg orally BID [4]. Another oral option that can be substituted for penicillin is amoxicillin at 20 mg/kg/day [8]. Monthly intramuscular injections of benzathine penicillin 600,000 units were used in older trials and have been used in other countries [36, 37]. This form of administration can be considered if compliance is an issue. Erythromycin is an option if the child is allergic to penicillin. The dose of erythromycin for those less than 3 years of age is 125 mg orally BID [36]. For those aged 3 to 5, the erythromycin dose is 250 mg orally BID. The various medications and dosages are listed in Table 1.

Table 1. Prophylactic Options for Infectious Caused by Streptococcus pneumoniae [4,5,8,36,37]

Medications	Age	Dose
Penicillin V potassium	< 3 years old	125 mg PO BID
Penicillin V potassium	≥ 3 years old	250 mg PO BID
Amoxicillin	No defined age	20 mg/kg/day PO
Benzathine Penicillin	No defined age	600,000 units IM monthly
Erythromycin	< 3 years old	125 mg PO BID
Erythromycin	≥ 3 years old	250 mg PO BID

BID = twice daily; IM = intramuscular; PO = oral

Another preventive measure for children with sickle cell disease is the pneumococcal vaccine. Since the advent of pneumococcal vaccination in 1977, the rates of invasive pneumococcal disease have markedly decreased among children [38]. Pneumococcal vaccination is therefore considered the mainstay of preventative therapy and is highly recommended by the Centers for Disease Control (CDC) and the Advisory Committee on Immunization Practices (ACIP) in the U.S. [39].

Produced in 1977, the 14-valent vaccine (PPV14) was one of the first vaccines developed for pneumococcal disease prevention. After a meta-analysis demonstrated that PPV14 failed to significantly reduce the risk of infection in children under three years of age, the pneumococcal polysaccharide vaccine (PPSV23) was brought to the market and replaced the 14-valent vaccine in 1983 [40]. The PPSV23 vaccine has been shown to protect against 80-90% of serotypes that cause invasive pneumococcal infections. However, since the polysaccharide vaccine provides little protective immunity in infants and young children, an effective vaccination is needed in children less than 3 years of age [41]. In 2000, the U.S. Federal Drug Administration (FDA) approved a 7-valent conjugate vaccine that accounted for 80-85% of invasive pneumococcal serotypes in infants and toddlers [42]. The Active Bacterial Core surveillance data, however, indicated that in 2008, a total of 61% of invasive pneumococcal disease cases among children aged <5 years were attributable to the serotypes covered in the currently-available 13-valent pneumococcal conjugate vaccine (PCV13), with serotype 19A accounting for 43% of the cases; PCV7 serotypes caused only <2% of the cases. In fact, three additional serotypes, (19A, 7F, and 3) accounted for 99% of the cases [43]. Broadening the coverage of the PCV7 vaccine serotypes was therefore highly desirable. On February 24, 2010, the FDA licensed a 13-valent pneumococcal conjugate vaccine. PCV13 is a pneumococcal conjugate vaccine that includes the 7 serotypes (4, 6B, 9V, 14, 18C, 19F, 23F) from PCV7 plus an additional six serotypes (1, 3, 5, 6A, 7F, 19A). Studies have shown that PCV13 is both safe and effective in preventing pneumococcal disease in children with SCD. In a study evaluating vaccine efficacy in high-risk patients, among those were children with SCD aged 6–18 years, 1 dose of PCV13 elicited significant immune response for all 13 serotypes as measured by serotype-specific IgG concentrations [44]. Because of this data, children with or without SCD should receive PCV13 at 2 months, 4 months, 6 months, and 12 months through 15 months of age.

Besides the PCV13 vaccine, children with SCD should also receive the PPSV23 vaccine. The pneumococcal polysaccharide vaccine was shown to be 80.4 % effective within 3 years after vaccination in pediatric patients with SCD (95% Cl 39.7 to 93.6; p <0.01) in

that they were less likely to be infected with pneumococcal serotypes that were in the vaccine [45]. Patients with SCD should receive the first PSSV23 vaccine at age 2 with at least an 8-week interval after the child's final dose of PCV13. These children should receive a second dose of PPSV23 five years after the first PPSV23 dose since the sustained protection of the vaccine over time is limited [46]. Together, these vaccines (i.e. PCV13 and PSSV23) can greatly cover most of the strains seen in invasive pneumococcal infections in both children and adults, especially in the SCD population [47].

Efficacy of penicillin prophylaxis

A Cochrane Review was recently published in 2012 that assessed the effects of prophylactic antibiotic regimens for preventing pneumococcal infections in children with sickle cell disease. Their objectives were also to explore the appropriate age that prophylactic penicillin could be discontinued and the adverse effects of penicillin from long-term prophylaxis. The review included three randomized, controlled trials comparing prophylactic antibiotics with placebo, no treatment or a comparator drug in preventing pneumococcal infection in children with sickle cell disease [6].

Selection criteria for trials considered for review were trials with children under the age of 16 years with homozygous sickle cell disease (HbSS), sickle beta thalassemia (Sβ0Thal and Sβ+Thal) and sickle cell hemoglobin SC. Primary outcomes included infections caused by S. pneumoniae and deaths. Secondary outcomes were adverse drug effects, antibiotic resistance, requirement for other course of antibiotics, and compliance with antibiotic prophylaxis [6].

Primary outcomes demonstrated a significant reduction (OR 0.37, 95%CI 0.16 to 0.86) of pneumococcal infection in children treated with penicillin. Overall, there were not any significant differences in the number of deaths among participants treated with penicillin prophylaxis and those who were not treated. The authors concluded that longer and larger trials are required to determine the impact on mortality [6].

For the secondary outcomes, minimal adverse effects of prophylactic therapy were seen and no significant differences in antibiotic resistant organisms were isolated. Endpoints such as the requirement for other antibiotics and compliance were unable to be accurately assessed due to the lack of data in the included trials. The authors in this Cochrane review concluded that penicillin prophylaxis reduced the incidence of pneumococcal infections in children with sickle cell disease under the age of 5 years and the risk of infection over 5 years of age was lower with no increase in the risk from discontinuing penicillin at this age [6]. These three trials in the Cochrane review validated the efficacy of antibiotic prophylaxis for pneumococcal infection in patients with SCD.

One of the first studies to look at the prevention of pneumococcal infection in children with SCD was by John et al in 1984 [37]. This trial looked at the efficacy of penicillin prophylaxis and the efficacy of the 14-valent pneumococcal vaccine in children aged six months to three years with homozygous sickle cell disease (SS) in preventing pneumococcal infection. Children could not be included if they had a documented pneumococcal infection or if they had undergone a splenectomy in the past. There were four different treatment groups: penicillin + 14-valent pneumococcal vaccine, 14-valent pneumococcal vaccine alone, penicillin + Haemophilus influenzae B vaccine, and Haemophilus influenzae B vaccine alone. The Haemophilus influenzae B vaccine was used as a control. The 14-valent pneumococcal vaccine contained antigens 1, 2, 3, 4, 6A, 7F, 8, 9N, 12F, 14, 18C, 19F, 23P, and 25. The penicillin was given intramuscularly as a long-acting benzathine penicillin at a dose of 600,000 units once a month [37].

Data was collected for a span of five years. During this time, there were 13 isolates of pneumococcus; 11 of these occurred in children who had received the pneumococcal vaccine. Only one serotype of the pneumococcus (type 36) was not included in the vaccine. The most common serotype isolated was type 23 which accounted for five cases. No pneumococcal isolates were seen in children receiving penicillin prophylaxis. After stopping penicillin prophylaxis, pneumococcal isolates were found in 7 children at 36 months of age. The most common adverse effect was transient pain occurring at the site of injection in the penicillin group. Due to this adverse reaction, penicillin prophylaxis was stopped at 36 months. This reaction also led to future trials using oral penicillin as a prophylactic option in children with SCD. This data suggested that pneumococcal vaccines given before two years of age did not guarantee protection against infection from pneumococcus; the authors suggested that vaccine boosters might be needed in the future. This trial confirms that penicillin offers the most

effective means of prophylaxis and should be continued past three years of age [37].

The PROPS trial published by Gaston et al [5] in 1986 guided the recommendations for penicillin prophylaxis in sickle cell patients. PROPS was a multicenter, randomized, double-blind trial conducted in the United States between August 1983 and June 1985. The purpose of the trial was to test whether the administration of penicillin twice a day would reduce the incidence of bacterial infection in a population of children with sickle cell anemia under the age of 3 [5].

Patients were included if they were between 3 to 36 months of age with hemoglobin SS. Patients were excluded if they were receiving long-term antibiotics or transfusion therapy. Patients were randomized to receive 125 mg of penicillin orally BID (n=105) or vitamin C 50 mg orally BID as placebo (n=110). The primary endpoint was documented severe infection of pneumonia. The secondary endpoint was infection due to an organism other than S. pneumoniae. Patients were seen at the clinic every three months. The clinic visits included a complete blood count, medical history, physical examination, a pill count, and urine collection to assess penicillin adherence. Blood and urine cultures were obtained when a child developed a severe infection in order to determine the presence of S. pneumoniae [5].

The study was terminated 8 months early due to an 84% reduction in S. pneumoniae infection in the penicillin group compared to the placebo group (p=0.0025). During the trial, 15 patients developed pneumococcal septicemia, 2 in the penicillin group and 13 in the placebo group. Infections occurred primarily in the younger children with 8 patients (53.3%) younger than 2 years of age, 5 patients (33.3%) who were 2 to 3 years of age, and 2 patients (13.3%) who were greater than 3 years of age. Four of the infected children who received placebo developed fulminant disease. Three of these patients proceeded from onset of fever to death in less than 9 hours. All three of these patients had received the 14-valent or 23-valent pneumococcal vaccine. The fourth child survived but had severe neurological impairment due to a cerebrovascular accident. No fatalities had occurred in the patients treated with penicillin. Compliance was hard to determine since urine samples were collected at only 31% of the scheduled return appointments and 34% of the appointments were missed. No allergic reactions were reported, and penicillin prophylaxis was well tolerated. The authors concluded that neonatal detection of sickle cell anemia should be a high priority and be the first step in the prevention of mortality and morbidity in patients with SCD. The authors recommended that babies identified at birth should receive penicillin no later than 4 months of age and that prophylaxis should continue beyond the third birthday [5].

After noting efficacy in the PROPS trial, the "Discontinuing penicillin prophylaxis in children with sickle cell anemia" trial, or the PROPS II trial, was conducted to determine the length of prophylaxis [19]. It is thought that older, school-aged children with sickle cell anemia appear less likely to develop pneumococcal bacteremia without penicillin prophylaxis. The adverse effects of penicillin prophylaxis past age 5 have not been well studied but are thought to increase the quantities of antibiotic-resistant strains of S. pneumoniae. The objective of this trial was to determine any adverse consequences of discontinuing penicillin prophylaxis in children with SCD after the age of 5 who had been on long-term penicillin prophylaxis [48].

Children with SS and Sβ0Thal were included if they had received penicillin prophylaxis for at least 2 years immediately before their 5th birthday and had received the 23-valent pneumococcal vaccine between their 2nd and 3rd year of life. Children could not have had a history of bacteremia or meningitis caused by S. pneumoniae or Hameophilus influenzae. Children were also excluded if they had a penicillin allergy, had a documented splenectomy, or were receiving long-term transfusions. They were randomized between ages 4 years and 9 months and 5 years and 3 months. During this time, they also received another immunization with the 23-valent pneumococcal vaccine; the child was not re-immunized if they had received the vaccine in the last year. Each child received 250 mg of penicillin V potassium orally BID or placebo orally BID. They were followed every 3 months in the clinic for a period of 3 years [48].

Throughout the trial, six children suffered from a systemic infection caused by S. pneumoniae. Of these, two were in the penicillin group (1.0%; 95% CI 0.1% to 3.6%) and four in the placebo group (2.0%; 95% CI 0.5% to 5.0%). Each of the isolates was either serotype 6A/6B or serotype 23F. These strains were tested for susceptibilities and found that four of the strains were penicillin susceptible and two of the strains were

Table 2. Comparison of clinical trials evaluating the efficacy of penicillin prophylaxis [5,37,48]

	John, et al, 1984	**Props, 1986**	**Props II, 1995**
Number of Patients	N = 242	N = 215	N = 400
Age Range	6 months – 3 years	3 months – 36 months	57 months – 6 3 months
Intervention	Benzathine penicillin 600,000 unit injecton + 14-valent pneumococcal vaccine, 14-valent pneumococcal vaccine alone, benzathine penicillin 600,000 unit injection + Hib vaccine, or Hib vaccine alone	Penicillin V potassium 125 mg BID or vitamin C 50 mg BID (placebo)	Penicillin V potassium 250 mg BID or placebo BID
Primary Outcome	Severe infection of pneumonia	Severe infection of pneumonia	Severe infection of pneumonia
Result	In the first 5 years, no pneumococcal infection in the penicillin group while receiving penicillin (4 isolates occurred 1 year after stopping); 11 infections in the pneumococcal vaccine group	84% reduction in *S. pneumoniae* infection (p=0.0025)	6 cases of infection: 4 in the placebo group (2.0%; 95% CI 0.5% to 5.0%); 2 in the penicillin group (1.0%; 95% CI 0.1% to 3.6%)

BID = twice daily; CI = confidence interval; Hib = Haemophilus influenzae type B; S. pneumoniae = Streptococcus pneumoniae

penicillin resistant. Five children suffered bacteremia caused by other organisms [H. influenzae type B and Salmonella in the patients who received penicillin; H influenzae type B (2) and group A β-hemolytic streptococcus in patients who received placebo]. This trial attempted to analyze a subset of patients for resistance by examining the presence of pneumococci in the nasopharynx. Of the 226 patients, about 25% were found to carry pneumococci in their nasopharynx at some point during the three-year study period [48]. It is, however, difficult to conclude if there is an increase in infections due to pathogens other than S. pneumoniae after a decrease in the carriage of the latter microorganism. However, most researchers do not think that the risk interactions within the nasopharynx will affect the prevalence of individuals' bacterial carriages [49].

The findings from PROPS suggested a risk of pneumococcal infection of 1.5 per 100 patient years in those under 3 years old receiving penicillin. In the age range of 4 to 5 years old, PROPS II demonstrated that only 0.67 per 100 patient years in the placebo group and half that in the penicillin group were at risk of pneumococcal infection [6]. These two trials outlined the decreased risk of pneumococcal infection with increased age. The authors thus concluded that children who had received comprehensive prophylactic care and those who had not had severe pneumococcal infection or a splenectomy could safely stop penicillin prophylactic therapy at age five [6]. There is still, however, a need for a more efficacious vaccine that covers all pneumococcal serotypes in order to provide protection for children with sickle cell disease [48]. Table 2 summarizes the major clinical trials that evaluated the efficacy of penicillin prophylaxis in pediatric patients with sickle cell disease.

Resistance of Streptococcus pneumoniae

Many reports have shown that drug-resistant S. pneumoniae has increased over the past decade [50]. An increased risk of penicillin-induced drug resistance in these patients may mean that as these children age, there will be less options of antibiotic treatment for potentially more severe infections. The risk of penicillin-induced drug-resistant S. pneumoniae, therefore, presents a valid concern during long-term prophylaxis of SCD patients and continues to be a

significant area of controversy. Moreover, patients with SCD have higher risk of developing resistant strain of S. pneumoniae than the general population [51].

Resistance of pneumococci lies in its cell wall. The pneumococcal cell wall plays an important physical barrier to the insertion of activated complement 5b and 9 into the cell membrane, which is also known as the membrane-attack complex. If this complex cannot go into the cell, then the organism will not be killed [52]. After being exposed to long-term penicillin therapy, there can be an alteration in the penicillin binding proteins of S. pneumoniae, mainly 1a, 1b, 2a, 2b, 2x, and 3. The changes to 1a, 2x, and 2b are most responsible for resistance of pneumococci (i.e. rising in the minimum inhibitory concentration) [53,54].

In the first PROPS trial, no resistant pneumococci were found in the nasopharynx of patients [5]. In a subset of patients in the PROPS II trial, 27% of the 226 patients were found to have S. pneumoniae. Nine percent of these patients had either penicillin intermediate or penicillin resistant isolates. There were not any significant differences between the groups treated and not treated with penicillin. There was, however, an increased likelihood for the children in the penicillin group to carry multiple drug resistant strains of pneumococci [48]. The clinical significance of these findings remains to be determined.

Another study by Sakhalkar et al [55] looked at the prevalence of penicillin-resistant strain of S. pneumoniae in pediatric SCD patients. Naso-pharyngeal cultures were compared between the SCD group and the control group. Six percent of the SCD patients had S. pneumoniae colonization, whereas 17% of the control group had such colonization. Among these isolates, 21% were resistant to penicillin, of which 50% were from the SCD group. The author concluded that penicillin prophylaxis helped reduce S. pneumoniae colonization in SCD patients, but it might increase the prevalence of penicillin-resistant strain of the organism.

Fonseca et al [24] evaluated the S. pneumoniae resistance pattern of the pediatric SCD patients in Sao Paulo, Brazil. Of the 14 strains of pneumococci that were isolated from the cohort, five of them (35.7%) showed resistance to oxacillin. Three of the five strains were considered as intermediate resistant to penicillin, with a mean inhibitory concentration (MIC) of 0.25 mcg/mL. No elevated resistant strains to penicillin were found in this study.

Strains of S. pneumoniae that are resistant to penicillin are increasing, and it is reported that around 40% of the isolates from SCD patients have some levels of resistance and most of these isolates can be found in patients on penicillin prophylaxis [56]. Penicillin prophylaxis is especially less effective in patients with intermediate-resistant and resistant serotypes. Vaccination with pneumococcal conjugate vaccine may help reduce the incidence of antibiotic-resistant pneumococcal infections [57].

McCavit et al [58] sought to describe the current practice of pediatric hematologists relating to stopping penicillin prophylaxis for SCD children after 5 years of age. One of the reasons that clinicians were comfortable in continuing the therapy past 5 years old was that they had decreased concern about antibiotic resistance to S. pneumoniae ($p = 0.01$). Even though some clinicians may not be concerned about resistance, the risk is there and such practice needs to be re-evaluated.

A study by Woods et al [29] looked at the influence of penicillin prophylaxis on antimicrobial resistance against S. pneumoniae in pediatric sickle cell patients. No difference was found among the 226 patients (penicillin group vs. placebo group) in the incidence of intermediate and resistant strains of pneumococcal colonization to penicillin. However, there was a trend toward increased incidence of multiple drug-resistant strains of S. pneumoniae in patients > 5 years old who had been receiving penicillin prophylaxis when compared to the control group. The author concluded that if the SCD patients did not have any history of invasive pneumococcal disease, clinicians should evaluate discontinuing the penicillin prophylactic therapy [29].

In the study mentioned earlier by Steele et al [30], over 50% of the pneumococcal strains that were isolated had some forms of resistance to penicillin, with 33% being intermediate (MIC of 0.06 to 1 mcg/mL) and 29% being resistant (MIC of >/= 2 mcg/mL). Such resistance was found to be associated with penicillin prophylaxis ($p < 0.01$). With the data presented above, penicillin prophylaxis in patients > 5 years old, especially those who have not had any invasive pneumococcal infection, should be re-evaluated to reduce the chance of developing resistance.

Resistance pattern of S. pneumoniae is different across the United States and the rest of the world. In adults, resistant strains of S. pneumoniae to penicillin were found in approximately 40% of infections that were

caused by this organism [50]. On the contrary, Donkor et al [59] found that the incidence of antibiotic resistance against S. pneumoniae was rather low in pediatric SCD patients in Ghana (0% with ampicillin, penicillin and cloxacillin and 11% with cefuroxime and erythromycin). Clotrimoxazole was the only antibiotic found to be extremely resistant to this strain of organism. At our institution, we had similar sensitivity pattern as in the study by Donkor et al [59], in which 100% of our isolates were sensitive to penicillin. Despite of the variability in the resistance pattern around the world, vancomycin seems to be the only commonly-used antibiotic that has yet developed any resistance [50].

Lastly, medication compliance may also play a role in the development of S. pneumoniae resistance [60]. Pradier et al [61] attempted to investigate penicillin's susceptibility to S. pneumoniae in 5 different European countries (i.e. France, United Kingdom, Spain, Germany, and Italy). The researchers found that higher prevalence rates of penicillin-resistant pneumococci were observed in Spain and France (i.e. 45% and 25%, respectively) than United Kingdom, Germany and Italy (i.e. 3%, 8%, and 5%, respectively) due to lower medications compliance rates in the former countries. The association of compliance with the development of S. pneumoniae resistance, however, is very difficult to capture because in most studies, compliance was unable to be accurately assessed due to the lack of data in the included trials [5,6].

Conclusion

The use of antibiotics like penicillin is effective in preventing pneumococcal infection in the pediatric population with SCD (i.e. SS and Sβ0Thal) less than 5 years old. Penicillin prophylaxis should be prescribed routinely in this population. Many of the clinical trials looking at vaccines only studied the 14-valent or the 23-valent vaccine. The new conjugate vaccine PCV13 may cover more of the serotypes seen with pneumococcal infections. The clinical impact of this new vaccine in addition to penicillin prophylaxis is unknown. It is important for these children to stay on schedule with their vaccinations, as this can help reduce the risk of infection along with penicillin prophylaxis. Fully immunized children who have not had a splenectomy or a severe pneumococcal infection and who have received comprehensive SCD care at a young age may be able to safely stop penicillin prophylaxis at 5 years old. It is important for caregivers to be educated and to be comfortable in seeking medical attention should the child develop a fever or become ill.

References:

1. Sickle Cell Disease Association of America, Inc. Sickle Cell Disease Global. April 12, 2014. Available at http://www.sicklecelldisease.org/index.cfm?page=scd=global. Accessed April 12, 2014.
2. DeBaun MR. Sickle Cell Disease. In: Kliegman RM, editor. Nelson Textbook of Pediatrics. 18th ed. Philadelphia, PA: Saunders; 2007.
3. John Hopkins Medicine. Sickle Cell Disease. April 12, 2014. Available at http://www.hopkinsmedicine.org/healthlibrary/conditions/hematology_and_blood_disorders/sickle_cell_disease_85,p00101/. Accessed April 12, 2014.
4. Adams, R., Billett, H. National Institutes of Health, National Heart, Lung and Blood Institute. Management of sickle cell disease. June 2002. Available at http://www. nhlbi.nih.gov/health/prof/blood/sickle/sc_mngt.pdf. Accessed April 12, 2014.
5. Gaston, H., Verter, J., Woods, G., et al., Prophylaxis with Oral Penicillin in Children with Sickle Cell Anemia. NEJM 1986; 314:1593-99.
6. Hirst C, Owusu-Ofori S. Prophylactic antibiotics for preventing pneumococcal infection in children with sickle cell disease. Cochrane Database Syst Rev. 2012 Sep 12.
7. Lin K, Barton MB. U.S. Preventive Services Task Force. Screening for Sickle Cell Disease in Newborns: U.S. Preventive Services Task Force Recommendation Statement. October 2014. Available at http://www.uspreventiveservicestaskforce.org/uspstf07/sicklecell/sicklers.htm. Accessed December 10, 2014.
8. American Academy of Pediatrics. Immunization in special clinical circumstances. In: Pickering LK, editor. Red Book: 2009 Report of the Committee on Infectious Diseases. 28th ed. Elk Grove Village, IL: American Academy of Pediatrics; 2009.
9. Pai VB, Nahata MC. Duration of penicillin prophylaxis in sickle cell anemia: issues and controversies. Pharmacotherapy 2000;20:110–117.
10. Infectious disease and Immunization Committee. Prevention and therapy of bacterial infections for children with asplenia or hyposplenia. Paediatr Child Health. 1999;4(6):417–31.
11. Brousse V, Buffet P, Rees D. The spleen and sickle cell disease: the sick(led) spleen 2014 Brit J of Haematol 166:165-76.
12. Andersson B,. Dahmen J, Frejd T, et al. Identification of an active disaccharide unit of a glycoconjugate receptor for pneumococci attaching to human pharyngeal epithelial cells. J. Exp. Med. 1983;158:559–70.

13. Boulnois GJ. Pneumococcal proteins and the pathogenesis of disease caused by Streptococcus pneumoniae. J. Gen. Microbiol. 1992;138:249–59.
14. Bloch EF, Castro O, Gregory JE, et al. Opsonization of pneumococci by whole serum from sickle cell disease patients. J Natl Med Assoc 1984;76-179-82.
15. Chudwin DS, Wara DW, Matthay KK, et al. Increased serum opsonic activity and antibody concentration in patients with sickle cell disease after pneumococcal polysaccharide immunization. J Pediatr 1983;102:51-4.
16. Bjornson AB, Lobel JS, Harr KS. Relation between serum opsonic activity for Streptococcus pneumoniae and complement function in sickle cell disease. J Infect Dis 1985;152:701-9.
17. Bjornson AB, Lobel JS. Lack of a requirement for the Fc region of IgG in restoring pneumococcal opsonization via the alternative complement pathway in sickle cell disease. J Infect Dis 1986;154:760-9.
18. Carddock PR. Bacterial infection in sickle-cell disease [letter]. N Engl J Med 1973;288:1301-2.
19. Dimitrov NV, Douwes FR, Bartolotta B, et al. Metabolic activity of polymorphonuclear leukocytes in sickle cell anemia. Acta Haematol 1972;47:283-91.
20. Rijkers GT, Sanders LA, Zegers BJ et al. Anti-capsular polysaccharide antibody deficiency states. Immunodeficiency. 1993;5(1):1-21.
21. AlonsoDeVelasco E, Verheul AF, Verhoff J, et al. Streptococcus pneumoniae: Virulence factors, pathogenesis, and vaccines. Microbiol Rev 1995;59(4): 591–603.
22. Wong WY, Overturf, GD, Powars DR. Infection caused by Streptococcus pneumoniae in children with sickle cell disease: epidemiology, immunologic mechanisms, prophylaxis, and vaccination. Clin Infect Dis 1992;14:1124-36.
23. Hord J, Byrd R, Stowe L, et al. Streptococcus pneumoniae sepsis and meningitis during the penicillin prophylaxis era in children with sickle cell disease. J Pediatr Hematol Oncol 2002;246:470-2.
24. Fonseca PB, Braga JA, Machado AM, et al. Nasopharyngeal colonization by Streptococcus Pneumoniae in children with sickle cell disease receiving penicillin prophylaixis. J Pediatr (Rio J) 2005;81(2):149-54.
25. Lloyd-Evans N, O.Dempsey TJ, Baldeh I, et al. Nasopharyngeal carriage of pneumococci in Gambian children and their families. Pediatr Infect Dis J. 1996;15:866-71.
26. Ferreira LLM, Carvalho ES, Berezin EN, et al. Colonização e resistência ntimicrobiana de Streptococcus pneumoniae isolado em nasofaringe de crianças com rinofaringite aguda. J Pediatr (Rio J). 2001;77:227-34.
27. Rey LC, Wolf B, Moreira LB, et al. S. pneumoniae isolados da nasofaringe de crianças sadias e com pneumonia: taxa de colonização e suscetibilidade aos antimicrobianos. J Pediatr (Rio J). 2002;78:105-12.
28. Battersby AJ, Knox-Macaulay HM, Carrol ED. Susceptibility to invasive bacterial infections in children with sickle cell disease. Pediatr Blood Cancer 2010;55:401-6.
29. Woods GM, Jorgensen JH, Waclawiw MA, et al. Influence of penicillin prophylaxis on antimicrobial resistance in nasopharyngeal S. pneumoniae among children with sickle cell anemia. The Ancillary Nasopharyngeal Culture Study of Prophylactic Penicillin Study II. J Pediatr Hematol Oncol 1997;19(4):327-33.
30. Steele RW, Warrier R, Unkel PJ, et al. Colonization with antibiotic-resistant Streptococcus pneumoniae in children with sickle cell disease. J Pediatr 1996;128:531-5.
31. Wiertsema SP, Kirkham LA, Corscadden KJ, et al. Predominance of nontypeable Haemophilus influenzae in children with otitis media following introduction of a 3+0 pneumococcal conjugate vaccine schedule. Vaccine. 2011;29(32):5163-70.
32. van Gils EJ, Hak E, Veenhoven RH, et al. Effect of seven-valent pneumococcal conjugate vaccine on Staphylococcus aureus colonisation in a randomised controlled trial. PLoS One. 2011; 6(6):e20229.
33. Dunne E, Manning J, Russell FM, et al. Effect of pneumococcal vaccination on nasopharyngeal carriage of Streptococcus pneumoniae, Haemophilus influenzae, Moraxella catarrhalis, and Staphylococcus aureus in Fijian children. J Clin Microbiol. 2012;50(3): 1034–8.
34. Morbidity and Mortality Weekly Report. Centers for Disease Control and Prevention. March 13, 1998. 47(09);169-172.
35. Ohene-Frempong K. Selected testing of newborns for sickle cell disease. Pediatrics 1989;83(5):879-880.
36. Knight-Madden J, Sergeant GR. Invasive pneumococcal disease in homozygous sickle cell disease: Jamaican experience 1973-1997. J Pediatr 2001;138:65-70.
37. John AB, Ramlal A, Jackson H, et al. Prevention of pneumococcal infection in children with homozygous sickle cell disease. Br Med J. 1984;288:1567-1570.
38. Centers for Disease Control and Prevention (CDC). Pneumonia hospitalizations among young children before and after introduction of pneumococcal conjugate vaccine—United States, 1997-2006. MMWR. 2009;58(1): 1-4.
39. Centers for Disease Control and Prevent (CDC). Licensure of a 13-valent pneumococcal conjugate vaccine (PCV13) and recommendations for use among children—Advisory Committee on Immunization Practices (ACIP), 2010. MMWR. 2009; 59(9): 258-61.
40. Davies EG, Hirst C, Lottenberg R, Dower N. Pneumococcal vaccines for sickle cell disease

(Review). Cochrane Database Syst Rev. 2004;(1):CD003885.
41. Lee CJ, Lee LH., Koizumi K. Polysaccharide Vaccines for Prevention of Encapsulated Bacterial Infections: Part 1. Infect Med. 2002;19(3):127-133.
42. National Network for Immunization Information. Pneumococcal. March 31, 2010. Available at http://www.immunizationinfo.org/vaccines/pneumococcal-disease. Accessed December 4, 2014.
43. Nuorti P., Whitney C. Prevention of Pneumococcal Disease Among Infants and Children- Use of 13-Valent Pneumococcal Conjugate Vaccine and 23-Valent Pneumococcal Polysaccharide Vaccine: Recommendations of the Advisory Committee on Immunization Practices. December 10, 2010/ 59 (RR 11):1-18.
44. Montalembert M, Abboud MR, Fiquet A, et al. A 2-dose schedule of 13-valent pneumococcal conjugate vaccine (PCV13) given to children with sickle cell disease previously immunized with 23-valent pneumococcal polysaccharide vaccine (PPSV23): results of a phase 3 study. Presented at the 54th Annual Meeting of the American Society of Hematology, Atlanta, GA; December 8–11, 2012.
45. Adamkiewicz TV, Sarnaik S, Buchanan GR, et al. Invasive pneumococcal infections in children with sickle cell disease in the era of penicillin prophylaxis, antibiotic resistance, and 23-valent pneumococcal polysaccharide vaccination. J Pediatr 2003;143(4):438-44.
46. Rao SP, Rajkumar K, Schiffman G, Desai N, Unger C, Miller ST. Antipneumococcal antibody levels three to seven years after first booster immunization in children with sickle cell disease, and after a second booster. J Pediatr 1995;127:590-2.
47. Center for Disease Control and Prevention. Pneumococcal Vaccination. September 18, 2014. Available at http://www.cdc.gov/vaccines/vpd-vac/pneumo /default. htm. Accessed December 15, 2014.
48. Falletta JM, Woods GM, Verter JI, et al. Discontinuing penicillin prophylaxis in children with sickle cell anemia. J Pediatr. 1995;127(5):685-90.
49. Adegbola RA, DeAntonio R, Hill PC, et al. Carriage of Streptococcus pneumoniae and other respiratory bacterial pathogens in low and lower-middle income countries: a systematic review and meta-analysis. PLoS ONE 2014: 9(8):e103293.
50. Billeter M; American Society of Health-System Pharmacists. ASHP therapeutic position statement on strategies for identifying and preventing pneumococcal resistance. Am J Health Syst Pharm. 2004;61(22):2430-35.
51. Jacobs MR. Streptococcus pneumoniae: epidemiology and patterns of resistance. Am J Med 2004;117 Suppl 3A:3S-15S.
52. Shin HS, Smith MR, Wood WB Jr. Heat labile opsonins to pneumococcus. II. Involvement of C3 and C5. J Exp Med 1969;130:1229-41.
53. Markiewicz Z, Tomasz A. Variation in penicillin-binding protein patterns of penicillin-resistant clinical isolates of pneumococci. J Clin Microbiol 1989;27:405-10.
54. Grebe T, Hakenback R. Penicillin-binding proteins 2b and 2x of Streptococcus pneumoniae are primary resistance determinants for different classes of ß-lactam antibiotics. Antimicrob Agents Chemother 1996;40:829-834.
55. Sakhalkar VS, Sarnaik SA, Asmar BI, et al. Prevalence of penicillin-nonsusceptible Streptococcus pneumonia in nasopharyngeal cultures from patients with sickle cell disease. South Med J 2001;94(4):401-4.
56. Steinberg MH. Pneumococcus and sickle cell disease: The beginning of the end? CID 2007;44:1434-5.
57. Kywa MH, Lynfield R, Schaffner W, et al. Effect of introduction of the pneumococcal conjugate vaccine on drug-resistant Streptococcus pneumoniae. N Eng J Med 2006;354:1455-63.
58. McCavit TL, Gilbert M, Buchanan GR. Prophylactic penicillin after 5 years of age in patients with sickle cell disease: a survey of sickle cell disease experts. Pediatr Blood Cancer 2013;60(6):935-9.
59. Donkor ES, Foster-Nyarko E, Enweronu-Laryea CC. Relationship between antibiotic resistance and sickle cell anemia: preliminary evidence from a pediatric carriage study in Ghana. Infect Drug Resistance 2013;6:71-7.
60. Pai VB, Nahata MC. Duration of penicillin prophylaxis in sickle cell anemia: issues and controversies. Pharmacotherapy 2000;20(1):110-7.
61. Pradier C, Dunais B, Carsenti-Etesse H, et al. Pneumococcal resistance patterns in Europe. Eur J Clin Microbiol Infect Dis 1997;16(9):644-7.