Asian Journal of Biotechnology and Bioresource Technology

2(4): 1-7, 2017; Article no.AJB2T.38835
ISSN: 2457-0125

Trends in Treatment and Vaccine Development of Typhoid Fever: A Review

M. Ali[1*], A. Yahaya[2], F. S. Nas[3], A. Anas[4] and I. S. Ibrahim[5]

[1]Department of Microbiology, Kano University of Science and Technology, Wudil, Nigeria.
[2]Department of Biology, Kano University of Science and Technology, Wudil, Nigeria.
[3]Department of Biological Science, Bayero University Kano, Kano, Nigeria.
[4]College of Basic and Remedial Studies, Hassan Usman Katsina Polytechnic, Katsina State, Nigeria.
[5]Department of Pharmaceutical Technology, School of Technology, Kano, Nigeria.

Authors' contributions

This work was carried out in collaboration between all authors. Authors MA, AY and FSN designed the study, wrote the protocol and wrote the first draft of the manuscript. Authors AA and ISI managed the analyses of the study. Author MA managed the literature searches. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/AJB2T/2017/38835
Editor(s):
(1) Sirigireddy Sivajothi, Professor, Department of Veterinary Parasitology, College of Veterinary Science, Sri Venkateswara Veterinary University, Andhra Pradesh, India.
Reviewers:
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(2) Akobi Oliver Adeyemi, Federal Medical Centre, Nigeria.
Complete Peer review History: http://prh.sdiarticle3.com/review-history/22909

Review Article

Received 16th October 2017
Accepted 7th January 2018
Published 29th January 2018

ABSTRACT

Typhoid fever is caused by bacterium called *Salmonella enterica* serovar *typhi* (often referred to as *S. typhi*). It has become rare developed countries, but remains an important cause of enteric disease in developing countries, resulting in an estimated 216,000 – 600,000 deaths per year, predominantly in children. Humans are the only source of infection of typhoid fever and transmission of *S. typhi* is by the fecal-oral route through contaminated water or food, prevention measures need to include water and sanitation improvements, as well as health education. Typhoid fever can be effectively treated with antibiotics, but growing rates of antibiotic resistance in many regions are making this treatment more difficult and costly. Given these facts, it seems necessary to consider a comprehensive approach to prevention of this disease that combines targeted vaccination as a short- to medium-term measure, combined with the long-term solutions

**Corresponding author: E-mail: alimuhd4real@gmail.com;*

of water and sanitation improvements and elevated living standards. To combat and to reduce the morbidity and mortality caused by typhoid fever, many preventive measures and strategies have been employed, the most important being vaccination. In recent years, many *Salmonella* vaccines have been developed including live attenuated as well as DNA vaccines and their clinical trials have shown encouraging results. However, with the increase in antibiotic resistance, the development of potent vaccine candidate for typhoid fever is needed.

Keywords: Typhoid fever; Salmonella typhi; vaccine; antibiotics.

1. INTRODUCTION

Typhoid fever is caused by a highly specific human adapted pathogen, Salmonella enterica serotype typhi. This organism is an important cause of febrile illness and death in population living in crowded and poorly sanitized environment. The risk of the disease is increasing in populations that are exposed to unsafe water and food and also pose a risk to travelers visiting to endemic country [1].

Salmonella typhi is a particular *Salmonella serovar* that causes typhoid fever, a major public health problem in developing countries [2]. Typhoid fever is a systemic disease in which the illness may last for three to four weeks and death rate range between 12% and 30%. Although the global burden of typhoid fever has been reduced, but the emergence of multidrug resistant *S. typhi* (MDRST) is still a threat to public health. *S. typhi* is a motile, facultative anaerobe bacterium that is susceptible to various antibiotics. Currently, 107 strains of this organism have been isolated; many containing varying metabolic characteristics, levels of virulence, and multi-drug resistance genes that complicate treatment in areas that resistance are prevalent [3]. Diagnostic identification can be attained by growth on MacConkey and Eosine Methylene Blue agars as the bacteria is strictly non-lactose fermenting. It also produces no gas when grown in Triple Sugar Iron agar, which is used to differentiate it from other Enterobacteriaceae [4].

Despite the emergence of newer antibacterial drugs, enteric fever has continued to be a major health problem [5]. *S. typhi* gained resistance to antibiotics like ampicillin, ceftriaxone, and cotrimoxazole, besides developing resistance to efficacious drugs like ciprofloxacin. The emergence of multidrug resistance to the commonly used antibiotics has further complicated the treatment and management of the disease and this is recognized as one of the greatest challenges in its management [6]. The importance of vaccination and other preventive measures for typhoid fever is heightened by increasing resistance of *Salmonella* serotype typhi to antimicrobial agents including fluoroquinolones in many parts of the world [5]. This paper aimed to review the current trends in treatment and vaccine development of typhoid fever.

2. TYPHOID FEVER

Typhoid fever is an infectious disease of global distribution [7]. It is a systemic infection caused by *Salmonella enterica* serotype *typhi,* and it is considered an important worldwide cause of morbidity and mortality [8]. It is a prolonged febrile illness and continues to be a health problem in developing countries where there is poor sanitation, poor standard of personal hygiene and prevalence of contaminated food. It is endemic in many parts of the developing world [9]. In urban areas where sewage disposal is lacking or inadequate, water supplies get contaminated and thus cause the outbreaks of typhoid. The contamination of food by carrier is the second most frequent route of infection [10]. A number of reports on typhoid were made in different part of the world [11] and a number of reports regarding the epidemiology of this disease have also been made [12].

2.1 *Salmonella typhi*

Salmonella typhi, the etiologic agent for typhoid fever, is gram negative bacteria belonging to the family Enterobacteriaceae. They are aerobic, non-spore-forming, flagellated bacilli of about 2-3 µm long and 0.4-0.6 µm diameter. Members of this genus have variety of pathogenic effect [13]. The strains have evolved to infect wide variety of reptiles, birds and mammals resulting in many different syndromes ranging from colonization and chronic carriage to acute fatal diseases. Differences in lipopolysaccharide (LPS) in *Salmonella* generate antigenic variations, which also affect the virulence of the strain [14].

S. typhi is generally transmitted via food or water contaminated with feces or urine from a case or

carrier. Direct person-to-person spread can also occur. Shellfish harvested from sewage-contaminated water are potential vehicles, as well as fruits and vegetables grown in soil fertilized with human waste in developing countries. Sexual transmission from an asymptomatic carrier has been documented. Laboratory-acquired infections also have been reported including laboratory workers who do not directly handle *Salmonella* specimens [13]. The emergence of multi drug resistance *S. typhi* (MDRST) has been of major concern in recent years. MDRST is defined as strains of *S. typhi* resistant to all three first line antibiotics for typhoid fever. The number of reported multi resistant typhoid fever increased rapidly throughout the world from 1989 onwards with most of the cases from the Africa, Middle East and Asia especially in the Indian subcontinent, Pakistan and China. Resistance to these agents is associated with the plasmid present in the bacteria [15].

2.2 Clinical Symptoms

The Initial clinical symptoms of typhoid fever include; sustained fever, abdominal pain, anorexia, lethargy, malaise, persistent headache, and a non productive cough. Constipation is reported more frequently than diarrhea in adults. Nausea and vomiting may also occur. Diarrhea is most common in children, especially infants under one year of age. During the second week of illness, there is often a protracted fever and mental dullness. Other symptoms may include intestinal bleeding, slight deafness, and parotitis. Neurologic symptoms which including acute psychosis, myelitis, meningitis, and encephalitis; focal central nervous infections occur rarely. Mild and typical infections are common and relapses occur in up to 15-20% of patients without treatment and symptoms may last for 3-4 weeks [16].

2.3 Pathogenesis

The infectious dose of *S. enteric* serotype typhi in patients varies between 1000 and 1 million organisms. Vi-negative strains of *S. enteric* serotype typhi are less infectious and less virulent than Vi-positive strains. *S. enteric* serotype typhi must survive the gastric acid barrier to reach the small intestine, and low gastric pH is an important defense mechanism. A chlorhydria as a result of aging, previous gastrectomy, treatment with proton-pump inhibitors or large amounts of antacids lowers the infective doses [17]. In the small intestine, the bacteria adhere to mucosal cells and then invade the mucosa. They rapidly penetrate the mucosal epithelium via either micro fold cells or enterocytes and arrive in the lamina propria, where they rapidly elicit an influx of macrophage that ingest the bacilli but do not generally kill them. Some bacilli remain within the macrophage of the small intestinal lymphoid tissue and some translocate to the intestinal lymphoid follicles and draining mesenteric lymph nodes by which they enter thoracic duct and general circulation [1]. As a result of the silent primary bacteraemia, the pathogens reach an intracellular haven within 24 hours after ingestion. *Salmonella typhi* are able to survive and multiply within mononuclear phagocytic cells of the lymphoid follicles, liver, spleen and bone marrow. The incubation period is usually 7 to 14 days. However, it depends on the number of bacteria, their virulence, and the host response. Clinical illness is accompanied by a fairly sustained level of secondary bacteraemia, usually one bacterium per millilitre of blood and about 10 bacteria per millilitre of bone marrow [18]. Typhoid fever induces systemic and local humoral and cellular immune responses, but these confer incomplete protection against relapse and re-infection. The interaction of host immunologic mediators and bacterial factors in infected tissue may contribute to necrosis of Peyer's patches in severe disease [19].

3. ANTIBIOTIC RESISTANCE IN *S. typhi*

The advent of antibiotic treatment has led to a change in the presentation of typhoid fever. However, rising of antibiotic resistance has been associated with increased severity of illness and related complications. A study was undertaken to compare the changing trends of antibiogram of *Salmonella enterica* serovar *typhi* and *Salmonella enteric* serovar *paratyphi A* isolates. A total of 80 isolates of *Salmonella* were obtained from blood cultures during 2001-2004. Identification and antibiotic sensitivity of the isolates were performed by using mini API (Bio Merieux, France). Sixty isolates were identified as *Salmonella enteric* serovar *typhi* and 20 were identified as *Salmonella enterica* serovar *paratyphi A*. More than 67% of *S. typhi* and 80% of *S. paratyphi A* isolates were sensitive to chloramphenicol. Sensitivity of *S. typhi* isolates to cephalosporins was found to have increased from 2001-2004 while that of *S. paratyphi A* showed a decline. With increasing resistance to ciprofloxacin and the possibility of re-emergence

of sensitivity to chloramphenicol, the policy of empirical treatment of enteric fever needs to be rationalized [20]. A total of 464 *Salmonella typhi* were isolated from blood of patients suspected with enteric fever in Calcutta school of medicine from 1991-2003. All the 464 isolates were susceptible to Amikacin and Gentamicin. Both antibiotics showed bactericidal activity at concentrations of 2 µg/ml respectively after incubation for 6 hrs. These drugs were administered for the treatment for typhoid fever [21]. Molecular characterization of *Salmonella typhi* with full resistance to ciprofloxacin and particularly the presence of plasmid borne integron in ciprofloxacin resistant *Salmonella typhi*, which will lead to a situation of untreatable enteric fever was first reported by Paul et al. [22].

In another work, all the *Salmonella Typhi* isolates were less sensitive to ciprofloxacin but no resistance was seen, whereas 76% of the same isolates showed resistant to nalidixic acid. In this work, it was shown that nalidixic acid resistant isolates had decreased susceptibility to ciprofloxacin [23]. Similar work was carried out by John et al. [24], which reveals same pattern of result as in the above. Changing trends of antibiogram of *S. typhi* and *S. paratyphi A* says that 67% of *S. typhi* in the study were sensitive to chloramphenicol and the sensitivity of *S. typhi* isolates to cephalosporin was found to have increased from 2001-2004 [20]. Resistance to amoxicillin, chloramphenicol, ampicillin and co-trimoxazole were significantly high in a study conducted in most part of the world [25]. Plasmid mediated multi-drug resistance to ampicillin, chloramphenicol and cotrimoxazole has been described in various parts of Asia [26].

3.1 Recent Trends of Drug Resistance and Its Effect in Treatment of Typhoid

Early diagnosis and prompt institution of appropriate antimicrobial are essential for optimal management. Knowledge of antibiotic susceptibility is crucial in determining which drug to use. More than 90% of patients can be managed at home with oral antibiotic and regular follow-up. However, patients with severe illness, persistent vomiting, severe diarrhoea, and abdominal distension, require hospitalization and parenteral antibiotic treatment [26]. Chloramphenicol was the drug of choice for several decades after its introduction in 1948. However, the emergence of plasmid mediate resistance and development of serious side effect like bone marrow aplasia had pushed this drug aside. Trimethoprim-sulfamethoxazole and ampicillin were employed to counter chloramphenicol resistance in 1970, but it was also discarded because of development of plasmid mediated resistance [27].

In the 1980s, ceftriaxone and ciprofloxacin became the drug of choice [27]. Although Fluoroquinolones attain excellent tissue penetration, rapid therapeutic response and very low rate of post treatment carriage, strains of bacteria have emerged in Asia that show resistance to them in the past decade [1]. Resistance to the fluoroquinolone may be total or partial. The nalidixic-acid-resistant strain has reduced susceptibility to fluroquinolone drug compared to nalidixic-acid-sensitive strain. Although isolates are nalidixic acid resistant but these can be susceptible to fluroquinolones in disc sensitivity testing [1]. The available fluroquinolones (Ofloxacin, Ciprofloxacin, Fleroxacin, perfloxacin) are highly active and equivalent inefficacy. For nalidixic-acid-resistant infections, a minimum of seven days of treatment at the maximum emitted dosage is necessary and 10-14 days are usually required. It is thought that Gatifloxacin is better than older fluoroquinolones. The bacteria need dual point mutations (in the DNA-gyrase and Topoisomerase-4genes) to became resistant to Gatifloxacin. Most studies in endemic countries have identified gyrA mutation in *S. enterica* as a mechanism of resistance [28].

Azithromycin in a dose of 500 mg (10mg/kg) given once daily for seven days has proven effective in the treatment of typhoid fever in adults and children. A dose of 1 g per day for five days was also found effective in adult. Of the third generation cephalosporin, oral Cefixime (15-20 mg per kg per day, for adults, 100-200 mg twice daily) has been widely used in children in a variety of geographical settings and found to be satisfactory. However, in some trials Cefixime showed higher rates of failure and relapse than fluroquinolones [1]. But antibiotic sensitivity pattern showed higher sensitivity around 78.8%. Intravenous third generation cephalosporins (Ceftriaxone,Cefixime, Cefotaxime) are effective with low relapse (3 to 6%) and fecal carriage (<3%) rates. Ceftriaxone is effectiveat a dose of 2-4 gm daily in single or two divided doses. Aztreonam and imipenem are potential third line drugs [18].

So, even in 2016, that is, 132 years after the isolation of the bacterium by the German

Scientist Gaffky, it is sad to say that typhoid fever is still an enigma. Every year throughout the world there are still around 15 million cases of typhoid out of which some 600,000 succumb to death [20]. Interestingly, more than 80% of all cases of typhoid fever belong to Asia and Africa. However, it is not uncommon in western countries either, like in the UK about 1 out of 1, 00,000 populations suffer from typhoid every year. Added to these problems, the treatment of typhoid fever has been even more challenging because of the emerging trends of resistant strains [22]. Microbial resistance regarding *Salmonella typhi* is basically of two types, viz., Quinolone Resistant *Salmonella typhi* (QRST), and multidrug resistant (MDR) type. There is also a strain which is known as DCS (decreased ciprofloxacin susceptibility) strain of *S. typhi* causing typhoid fever [22,23].

A study on knowledge, attitude and practice of general practitioners (GP) regarding treatment of typhoid fever by Paul et al. in January 2016, it has shown that the antibiotics used by GPs for treatment of typhoid are: Azithromycin (42%), Fluoroquinolones (32%), Cefixime(16%), Amoxycillin (6%) and Choramphenicol (4%). A combination of antibiotics is preferred by 38% of GPs and the preferred combinations of antibiotics are Cefixime + Azithromycin (26%) and Ciprofloxacin + Azithromycin (12%). Parenteral antibiotic is preferred in most cases when the patient is unable to consume orally usually due to excessive vomiting. Thus, a new plan and expanded thinking is now required regarding present and future management of typhoid fever [22].

4. VACCINE DEVELOPMENT: CURRENT STATUS

The first parental whole-cell typhoid vaccine was introduced in 1896 and used in England and Germany but withdrawn by most of the countries because of strong side effects [29]. The live oral vaccine Ty21a is available in enteric-coated capsule or liquid formulation. It should be taken in three doses two days apart on an empty stomach and is suitable for adult and children at least 5 years. The vaccine is well tolerated. Because it is live attenuated, it should not be given in immune compromised and in patients who is taking antibiotic [30]. The parenteral Vi-based vaccine is suitable for adult and children over the age of two years and a single dose of 0.5 ml is administered intramuscularly. In field trials conducted in Nepal and South Africa, where Typhoid is endemic, the protective efficacy of the vaccine was 72%, one and half years after vaccination. Booster dose is recommended every two years [30]. The WHO, recommends vaccination for people travelling in high-risk areas where the disease is endemic, people living in such areas, people in refugee camps, microbiologists, sewage workers and children [29].

4.1 DNA Vaccine for Typhoid Fever

Epitope selection based vaccine development has been tried for long. The designing of the small DNA sequence based vaccine for many infectious diseases such as typhoid fever seems to be a promising area of research. Many efforts are being carried out in this direction. In 2004, a report stated that certain antigens of *Salmonella* showing high expression *in vivo* were preferentially recognized by CD4 T cells. Five peptides (Mig-14, licA, SseB, SsaJ, or SifB) of *Salmonella* were selected on the basis of maximum *in vivo* expression. These peptides were GFP tagged and selected out from many proteins tested for the *in vivo* expression profile. The studies of typhoid fever vaccination showed that SseB and Mig-14 were exceptionally efficacious antigens providing antigen specific immunity and protection as compared to other antigens used. Other than high expression level, there are certain antigenic parameters which can influence protective efficacy and show different immune response for different antigen [28]. In another DNA vaccine study sopB protein of *Salmonella* was chosen based on its ability to induce better cell mediated immunity. The vaccine was able to confer protection against *Salmonella* challenge (lethal dose). The protection and immunity was further enhanced when used in combination with live attenuated vaccine candidates [7].

4.2 Need for New and Effective Vaccine for Typhoid Fever

The development of a new vaccine for typhoid fever is a challenge for scientists to pursue. Several vaccines have undergone clinical trials but public acceptance is still required. The need of an efficacious and potent vaccine against typhoid fever which can be used in children below 2-5 yr of age providing strong humoral as well as cellular immunity still persists.

5. CONCLUSION

Enteric fever remains a major public health challenge. Although in various parts of the world,

azithromycin show promise for the management of fever, till now, ceftriaxone has proven as most effective drug, precaution should be taken about serious side effect like hypersensitivity during administration. Now-a-days life threatening effects like cardiotoxity is also reported. Historical adaptation of *Salmonella* to patterns of antimicrobial use suggests the vigilance for the emergence of ceftriaxone-resistant strains. Indiscriminate and injudicious use of antibiotics raises the risk of newer strains of resistant organisms. Uses of antibiotic should be guided by culture and sensitivity report, over-the-counter sales of antibiotics must be stopped to prevent this nightmare.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
The peer review history for this paper can be accessed here:
http://prh.sdiarticle3.com/review-history/22909