Clinica Chimica Acta xxx (2015) xxx–xxx

Contents lists available at ScienceDirect

Clinica Chimica Acta

journal homepage: www.elsevier.com/locate/clinchim

Invited critical review

Thimerosal: Clinical, epidemiologic and biochemical studies ☆

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ARTICLE INFO

Article history:
Received 9 January 2015
Received in revised form 6 February 2015
Accepted 14 February 2015
Available online xxxx

Keywords:
Thimerosal
Ethylmercury
Humans
Neurodevelopment

ABSTRACT

Introduction: Thimerosal (or Thiomersal) is a trade name for an organomercurial compound (sodium ethylmercury (Hg) thiosalicylate) that is 49.55% Hg by weight, which rapidly decomposes in aqueous saline solutions into ethyl-Hg hydroxide and ethyl-Hg chloride. Developed in 1927, it has been and is still being used as a preservative in some cosmetics, topical pharmaceuticals, and biological drug products, including vaccines. Concerns have been voiced about its use because it is toxic to human cells. Although it is banned in several countries, it continues to be added to some vaccines in the United States and many vaccines in the developing world.
Discussion: This critical review focuses on the clinical, epidemiological, and biochemical studies of adverse effects from Thimerosal in developing humans. This review will include research that examines fetal, infant, and childhood death; birth defects; neurodevelopmental testing deficits in children; and neurodevelopmental disorders (attention deficit/hyperactivity disorder, autism spectrum disorder, tic disorder, and specific developmental delays). The review will also look at the research that examined the outcomes of acute accidental ethyl-Hg poisoning in humans. The studies that examine the underlying biochemical insights into the neuronal cellular damage will also be explored.
Conclusion: The culmination of the research that examines the effects of Thimerosal in humans indicates that it is a poison at minute levels with a plethora of deleterious consequences, even at the levels currently administered in vaccines.

Contents

☆ Conflict of interest: Six (6) of authors have been involved in vaccine/biologic legal actions (DAG, PGK, BSH, JKK, LSK, and MRG). One author (JGD) has no conflict of interest.

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http://dx.doi.org/10.1016/j.cca.2015.02.030
0009-8981/

Please cite this article as: Geier DA, et al, Thimerosal: Clinical, epidemiologic and biochemical studies, Clin Chim Acta (2015), http://dx.doi.org/10.1016/j.cca.2015.02.030

1. Introduction

Thimerosal (or Thiomersal) is a trade name for an organomercurial compound (sodium ethyl-mercury (Hg) thiosalicylate, $C_9H_9HgNaO_2S$) that is 49.55% Hg by weight. Thimerosal quickly decomposes in aqueous saline solutions into ethyl-Hg hydroxide and ethyl-Hg chloride [1]. Developed in 1927, it has been used as a preservative in cosmetics, pharmaceutical preparations, and biological products such as eye shadows, make-up removers, mascaras, and soap-free cleansers (cosmetic products); ear, eye and nose drops and ointments, antiseptic sprays, topical medications and tincture of Merthiolate (pharmaceutical preparations); and antitoxins, immune globulin preparations, skin-prick test antigens, and vaccines (biological products) [2].

Hg compounds have been used as disinfectants since bacteriology began [3]. For a long period of time, Hg compounds, such as mercury chloride ($HgCl_2$), were thought to be useful in the killing of bacteria and other microorganisms [3]. Despite this fact, as early as 1943, it was reported that plasma preserved with 1:10,000 Thimerosal was contaminated with viable micro-organisms, and it was concluded that Thimerosal cannot be considered the ideal preservative [4]. Subsequently, Morton et al. [3] reported that the label for Thimerosal (solution of 1:1000) stated that Thimerosal is a stainless and stable organic mercury compound of high germicidal value, especially in serum and other protein media. However, Morton et al. [3], based upon their experiments, found that Thimerosal is not highly germicidal and does not possess high germicidal value in the presence of serum and other protein mediums particularly. They further stated that the loss of antibacterial activity of mercurials in the presence of serum proves their incompatibility with serum. Furthermore, these investigators described that Thimerosal was 35-times more toxic to embryonic tissue cells than it was to bacteria, as well as more toxic to leukocytes than bacteria [3].

In more recent research, the effectiveness of Thimerosal as a preservative in Diphtheria–Tetanus–Pertussis (DTP) vaccine was evaluated by the United States (US) Centers for Disease Control and Prevention (CDC) [5]. The CDC reported that the choice and level of the preservative for inclusion in DTP vaccine were limited because of possible harmful effects on the vaccine's antigenicity, plus the need to ensure safety of the preservative. These investigators reported that Thimerosal, the preservative used in the production of DTP as an organic-Hg bacteriostatic agent, was only weakly bactericidal. The laboratory experiments revealed up to 2 week survival of bacterial cells in multi-dose DTP vaccine vials using Thimerosal as a preservative. These investigators concluded that at currently used concentrations, Thimerosal is not an ideal preservative. Higher concentrations were not recommended because it might reduce vaccine potency or pose a danger to individuals receiving the vaccine. As a result, the investigators suggested that those administering Thimerosal preserved vaccines should not rely on its effectiveness, but instead should apply particular attention to sterile technique when using multi-dose vials. Other investigators observed that Thimerosal failed to meet European Pharmacopoeia (EP) antimicrobial effectiveness acceptance criteria as a preservative due to lack of growth inhibition of Thimerosal on *Staphylococcus aureus* in both single and multi-challenge evaluations [6]. Finally, other investigators described the toxicity levels of commonly used preservatives in vaccines and biologics [7]. When comparing the relative cytotoxicity levels of the preservatives in US licensed vaccines, the observed relative toxicity of the compounds tested was phenol < 2-phenoxyethanol < benzethonium chloride < Thimerosal, and the relative toxicity indices (human neuroblastoma cells/bacterial cells) were 2-phenoxyethanol (4.6-fold) < phenol (12.2-fold) < Thimerosal (>330-fold). For the products tested, except for 2-phenoxethanol, the amounts needed to cause significant killing of bacteria were much higher than those routinely used in US licensed vaccine/biological preparations.

Despite all of the aforementioned concerns and the fact that there are other approved and effective preservatives available [6,7], Thimerosal continues to be used as a preservative in several vaccines to date and is a considerable source of Hg exposure for children [8,9]. About 50% of the Hg exposure in infants comes from the recurring bolus doses of Thimerosal from Thimerosal-containing vaccines administered in the first 2 years of life (cumulative doses of Hg exposure from Thimerosal-containing vaccines can be as high as 187.5 μg Hg in the first six months of life) [9]. Although this degree of exposure in the first six months of life has been reduced in the US in recent years, it remains unchanged in developing countries. There is considerable body of scientific and medical evidence supporting a role from Hg exposure causing harmful consequences [10]. To date, there are at least 180 studies that show harm from Thimerosal [11]. The purpose of this review is to specifically examine human clinical, epidemiological, and biochemical studies demonstrating the developmental adverse affects from human exposure to Thimerosal and its ethyl-Hg breakdown products.

2. Thimerosal exposure from vaccines

Until the beginning of this century, every tetanus-containing vaccine in the US (e.g., the DTP, tetanus toxoid (TT), diphtheria–tetanus (DT), and diphtheria–tetanus–acellular-pertussis (DTaP)), Haemophilus influenza type b (Hib), hepatitis B (HepB), and a polysaccharide meningococcal meningitis A, C, Y, and W-135 vaccine contained Thimerosal, many at a concentration of 0.01% Thimerosal. However, on July 7, 1999, the US Public Health Service (USPHS) and American Academy of Pediatrics (AAP) called for the elimination of Thimerosal from all vaccines in the US as soon as possible [12]. Then, as the vaccines were approved by the US Food and Drug Administration (FDA), reduced-Thimerosal vaccines began to displace the previous Thimerosal-preserved vaccines in the early 2000s. Finally, beginning in the late 2000s, no-Thimerosal vaccines began to replace the reduced-Thimerosal vaccines in the US. However, to date, the US FDA has not canceled the licenses for the Thimerosal-preserved vaccines or kept them from being produced and marketed [13].

As more of the reduced-Thimerosal and no-Thimerosal vaccines became available in the early 2000s in the US, the assumption was that the exposure to Thimerosal would sharply decrease. However, this expectation proved to not be accurate because of recommendation changes in the vaccination schedule. Starting in April of 2002, the US CDC began to recommend that influenza vaccines be given to infants and children, who were 6-to-23 months of age, when the only approved influenza vaccine for that age group was preserved with Thimerosal (Sanofi Pasteur's Fluzone®). In addition, the US CDC recommended influenza vaccines be given to women who were pregnant in their second and third trimesters, when the available influenza vaccines were also Thimerosal preserved [14]. In addition, through 2010, the US CDC progressively widened the age range for annual influenza vaccine such that very young children were supposed to get two doses of influenza vaccine initially (at 6 and 7 months of age) and then receive an additional dose every year. By this time, the US CDC had also discontinued the "second-and-third-trimester" constraint on giving influenza vaccines to pregnant women [15–17].

Thus, even though the US FDA eventually approved the reduced-Thimerosal and no-Thimerosal formulations of the tetanus-containing vaccines and some other vaccines, exposure to Thimerosal through vaccination has remained common in the US. As recently as 2013, more than half of all the influenza vaccines were still preserved with Thimerosal. Therefore, the approximate maximum lifetime exposure to Hg from Thimerosal-preserved vaccines has increased compared to the lifetime exposure under the US CDC's pre-2000 recommended vaccination schedule. It is estimated that it is now more than double what it would have been had the pre-2000 vaccination schedule been maintained. To date, in the US, Thimerosal is still a preservative in some of the other US FDA-approved vaccines including a multi-dose tetanus toxoid (TT) vaccine, and one multi-dose meningococcal meningitis vaccine [18]. Estimations suggest that there has not been a major decrease in Hg exposure from Thimerosal-preserved vaccines in vaccine-schedule-compliant children in the US.

Please cite this article as: Geier DA, et al, Thimerosal: Clinical, epidemiologic and biochemical studies, Clin Chim Acta (2015), http://dx.doi.org/10.1016/j.cca.2015.02.030