



ACIP

2025

## Thimerosal as a Vaccine Preservative

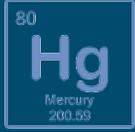
Prepared for the  
Advisory Committee for  
Immunization Practices

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# The Food and Drug Administration Modernization Act of 1997

- Compiled list of drugs and foods that contained intentionally introduced mercury compounds
- Provided quantitative and qualitative analysis of mercury compounds
- Report included 219 products which included 11 biologics and immunoglobulin therapies



## 1999 Joint Statement by US Public Health Service (PHS) and American Academy of Pediatrics (AAP)

- On July 9th, 1999, a joint statement issued by PHS and AAP called for the immediate reduction and elimination of the mercury based preservative thimerosal from infant vaccines based on the findings that infants and children who received vaccines preserved with thimerosal **could be exposed to mercury in excess of federal safety guidelines.**
- “because any potential risk is of concern, the Public Health Service (PHS), the American Academy of Pediatrics (AAP), and vaccine manufacturers agree that thimerosal-containing vaccines should be removed as soon as possible.”

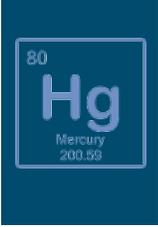


## Institute of Medicine

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# Immunization Safety Review of Thimerosal Containing Vaccines and Neurodevelopmental Disorders (October 2001)

- The committee “supported prior decisions made by the ACIP and AAP to call for the removal of thimerosal from vaccines that are part of the recommended childhood immunization schedule.” But they noted that “vaccines that are not part of the recommended childhood immunization schedule still contain thimerosal as a preservative and may be given to some children” and a lingering concern that there remains “on the shelf” an unknown quantity of thimerosal-containing Hib, hepatitis B, and DTaP vaccines.
- Recommended: “the use of thimerosal free DTaP, HiB and Hepatitis B vaccines despite the fact that there might be remaining supplies available” and “full consideration be given by appropriate professional societies and government agencies to removing thimerosal from vaccines administered to infants, children or pregnant women in the united States.”



# Safety Studies

## Prior to Marketing Thimerosal as Vaccine Preservative

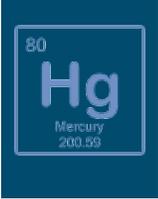
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- In 1997, FDA Modernization Act prompted assessment of thimerosal use in vaccines.
- FDA was unable to find any clinical studies formally evaluating the use of thimerosal before its initial marketing in 1930's. (1)
- Single study published in 1931 where thimerosal administered to individuals suffering from meningitis. (2)
  - Not designed to specifically examine toxicity and no clinical assessments or laboratory studies reported.
- FDA grandfathered in the use of thimerosal and did not require the typical animal safety data for finished biological products, including active and inactive ingredients. (3)

(1) Ball LK, Ball R, Pratt RD. An assessment of thimerosal use in childhood vaccines *Pediatrics*. 2001;(107)5:1147-1153.

(2) Powell HM, Jamieson WA. Merthiolate as a germicide. *Am J. Hyg.* 1931;(13):296-310.

(3) Department of Health and Human Services. 21 Code of Federal Regulations: 601.25. 1985



# Thimerosal use in vaccines

- Thimerosal was first introduced as a preservative in vaccines in the 1930's after being developed and patented in 1927 and subsequently marketed by Eli Lilly under the trade name "Merthiolate" in 1928.
- After a deadly incident of bacterial contamination of vaccines in Australia where 12 children died from staphylococcal-contaminated diphtheria vaccine, the FDA began enforcing safety standards which led to regulations requiring that all multidose vaccines contain a preservative to prevent contamination.
- Thimerosal was used during the manufacturing process and in multi-dose vials to prevent bacterial and fungal contamination. By the 1930's thimerosal became widely used as a preservative in vaccines and other medical products.



# Thimerosal Not Generally Recognized as Safe and Effective (GRASE) in OTC Drugs

- In 1975, FDA convened panel of experts to evaluate the use of mercury-containing over-the-counter (OTC). The panel found evidence that thimerosal was “35.3 times more toxic for embryonic chick heart tissue than for *Staphylococcus aureus*” and “was no better than water in protecting mice from potential fatal streptococcal infections.”
- The FDA issued a report of the panel’s findings in the Federal Register in 1982 which concluded that “thimerosal was not safe for OTC topical use because of its potential for cell damage if applied to broken skin and its allergy potential, not effective “because bacteriostatic action could be reversed.”
- In response to the expert panels report, the FDA published final rules in the Federal Register in 1998 that concluded **the use of thimerosal in over-the-counter products is not “generally recognized as safe or effective.” (GRASE)**

Ref: Federal Register, Department of Health and Human Services, Food and Drug Administration. *Mercury-Containing Drug Products for Topical Antimicrobial Over-the-Counter Human Use; Establishment of a Monograph*. (January 5, 1982);47(2):436-442. 47 FR 436 [Docket No. 75N-0183(1)]  
Federal Register, Department of Health and Human Services, Food and Drug Administration. *Status of Certain Additional Over-the-Counter Drug Category II and III Active Ingredients*. (April 22, 1998);63(77):19799-19802. 21 CFR Part 310 [Docket No. 75N-183F, 75N-183D, and 80N-0280].



# Evidence of Thimerosal Ineffectiveness as a Preservative

- In 1982, clusters of disease from Group A streptococcus infections were traced back to multi-dose vials of diphtheria toxoid, pertussis, and tetanus toxoid (DPT) vaccine which were contaminated after being opened. Thimerosal was present in acceptable limits in unopen vials of vaccine from the same lot. The authors concluded that **“Preservatives in multidose vials do not prevent short term bacterial contamination”** and the **“only feasible and cost-effective preventative measure now available is careful attention to sterile technique when administering vaccines from multidose vials.”**
- In 2004, a Chiron plant that manufactured Fluvirin vaccine preserved with thimerosal was forced to close because its vaccine was contaminated with *Serratia marcescens*. The closure created shortages in the vaccine supply and caused concern among providers and patients. In this case and others, thimerosal failed to prevent bacterial growth.

Stetler HC, Garbe PL, Dwyer DM, Facklam RR, Orenstein WA, West GR, Dudley KJ, Bloch AB. Outbreaks of group A streptococcal abscesses following diphtheria-tetanus toxoid-pertussis vaccination. *Pediatrics*. 1985 Feb;75(2):299-303. PMID: 3881728.

Smith S. Safety fears cut vaccine for flu. Priority urged for the aged, frail. *Boston Globe*. (October 6, 2004).

[http://www.boston.com/news/globe/health\\_science/articles/2004/10/06/safety\\_fears\\_cut\\_vaccine\\_for\\_flu?mode=PF](http://www.boston.com/news/globe/health_science/articles/2004/10/06/safety_fears_cut_vaccine_for_flu?mode=PF)



# Current Research

## Thimerosal safety and effectiveness

- A more current in vitro investigation into the cytotoxic effects and antimicrobial activity of thimerosal was published in 2023. The authors reported that thimerosal **should be present in culture media at 100 µg/ml** concentration to achieve effective antimicrobial activity. **But all tested cell lines lost viability completely at 4.6 µg/mL.**
- “Overall, our study revealed **Thimerosal was 333-fold more cytotoxic to human and animal cells as compared to bacterial and fungal cells.** Our results promote more study on Thimerosal toxicity and its antimicrobial effectiveness to obtain more safe concentrations in biopharmaceuticals.”



# Evidence of Thimerosal Developmental Neurotoxicity

- In 1987 the Commission of the European Communities initiated a research project on 10 known or suspected spindle poisons. Thimerosal was found to cause significant interference with microtubule polymerization destabilizing the spindle machinery that ensures accurate chromosome separation during cell division.
- This mechanistic disruption suggests direct mutagenic potential via structural chromosomal abnormalities, which is a known pathway to carcinogenesis or congenital defects.
- By affecting fundamental cellular structures at the molecular level, thimerosal revealed **“strong mechanistic evidence for mutagenicity and developmental toxicity”**.

Wallin, M. (1993). Effects of potential aneuploidy-inducing agents on microtubule assembly in vitro.

\*Mutation Research. Fundamental and Molecular Mechanisms of Mutagenesis\*, 287(1), 17–22. [https://doi.org/10.1016/0027-5107\(93\)90141-2](https://doi.org/10.1016/0027-5107(93)90141-2)



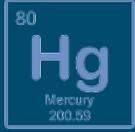
# CALIFORNIA EPA Proposition - 65

- California's Proposition 65, requires identification of chemicals known to cause cancer, birth defects or other reproductive harm, and to ensure public warnings when people might be exposed to them. Thimerosal has been on the Prop-65 list since 1990.
- In 2003 a petition was filed on behalf of the Bayer Corporation "for reconsideration of the determination that mercury and mercury compounds (thimerosal) as reproductive toxicants."
- The CA EPA responded that "The scientific evidence that thimerosal cause reproductive toxicity is clear and voluminous. Thimerosal dissociates in the body to ethyl mercury. **The evidence for its reproductive toxicity includes severe mental retardation or malformations in human offspring who were poisoned when their mothers were exposed to ethyl mercury or thimerosal while pregnant**, studies in animals demonstrating developmental toxicity after exposure to either ethyl mercury or thimerosal, and data showing interconversion to other forms of mercury that also clearly cause reproductive toxicity."



# Evidence From Human Research (1)

- A study published in Pediatrics in 2000 measured blood mercury levels in preterm and term infants after administration of the Hepatitis B vaccine containing 12.5 µg ethyl mercury.
- The investigation documented elevated post-immunization concentrations relative to pre-immunization levels in all neonates studied. Levels of blood mercury after exposure in low-birth-weight infants were 7.36 ( $\pm$  4.99) µg/L.
- One infant was found to have developed a mercury level of 23.6 µg/L, thus meeting the CDC criteria as a case of chemical poisoning from mercury 10µg /L. The study subjects had measurable blood Hg concentrations prior to immunization, indicating that risk assessment must include background mercury levels from other sources.



## Evidence From Human Research (2)

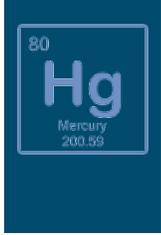
- Experts contend that there are “windows of vulnerability” which occur during neurological development and that specific types of developmental outcomes may have separate windows of vulnerability. These critical periods of development have not been established and may be relatively short in duration.
- The fact that thimerosal from vaccines has been documented to raise blood mercury levels over known thresholds where developmental effects have been documented to occur during the first few months of life, means that particular "windows of vulnerability" may have been breached. Even minor neurological impairment can have profound societal effects when amortized across the entire population and life span.



# Exposure To Vaccine Level Thimerosal Crosses the Blood Brain Barrier and Results in Significant Deposition of Mercury in the Brain

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- A 2005 study funded by NIH compared brain mercury levels in infant *Macaca fascicularis* primates exposed to: 1) injected ethyl mercury (thimerosal) and 2) equal amounts of ingested methylmercury.
- Ethyl mercury more rapidly converted to inorganic mercury in the brains of the primates which resulted in increasing levels of inorganic mercury.
- Primates exposed to ethyl mercury retained much higher levels of inorganic mercury in their brains, up to 71% vs. 10% compared to primates exposed to methyl mercury.
- Once organic mercury compounds reach the brain tissue and dealkylate,  $\text{Hg}^{2+}$  (toxic inorganic mercury) the mercury cannot cross the blood brain barrier and becomes trapped, resulting in neuroinflammation.

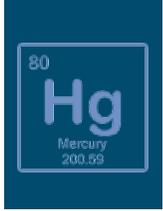


# Inorganic Mercury and the Developing Brain | 15

- A recent review outlined evidence from human and animal studies which estimated the inorganic mercury half-life in human brains of at least five to 27 years. (1)
- The impact of mercury exposure to the developing brain interferes with neuronal proliferation, migration, differentiation, synaptogenesis, tightly regulated apoptosis, and other processes vital to the formation and functioning of the nervous system which can result in lifelong neurodevelopmental impacts.
- Exposure during the first trimester of pregnancy may result in deficits or defects very different from those developed by someone who is exposed during the third trimester of pregnancy. **“There may never be a safe level of mercury exposure, especially for an unborn child.” (2)**

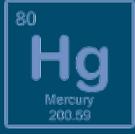
1. Rooney JP. The retention time of inorganic mercury in the brain--a systematic review of the evidence. *Toxicol Appl Pharmacol*. 2014 Feb 1;274(3):425-35.

2. Pletz J, Sánchez-Bayo F, Tennekés HA. Dose-response analysis indicating time-dependent neurotoxicity caused by organic and inorganic mercury-Implications for toxic effects in the developing brain. *Toxicology*. 2016 Mar 10;347-349:1-5. doi: 10.1016/j.tox.2016.02.006. Epub 2016 Mar 2. PMID: 26945727.



# Mercury Content In Thimerosal-containing Flu Vaccines

- Thimerosal is ~49.6% mercury by weight with a concentration of 0.01% thimerosal in vaccines. This equates to 100 µg/mL of thimerosal and 50 µg/mL of ethyl mercury per ml.
- The EPA's Toxicity Characteristic Leaching Procedure (TCLP) determines whether a substance is considered hazardous waste based on its potential to leach toxic chemicals into groundwater.
- According to EPA TCLP Mercury Threshold (40 CFR § 261.24) the regulatory threshold for mercury (D009) under TCLP is: 0.2 mg/L (200 parts per billion, or 200 µg/L).
- Flu vaccine mercury concentration (~50,000 µg/L) is 250 times greater than the TCLP limit. **Therefore, thimerosal-containing vaccines exceed the TCLP threshold by orders of magnitude and are classified as D009 Hazardous Waste**



# Vaccine Mercury: Disposal Guidelines



## PharmEcology® Provides Disposal Guidelines for the 2024-2025 Flu Season

PharmEcology Services, a division of WM Sustainability Services, is pleased to publish a summary of the proper flu vaccine disposal guidelines for the 2024-2025 flu season as a public service to the healthcare community. Please also see additional information regarding this year's vaccines following the table below.

### Flu vaccine summary table:

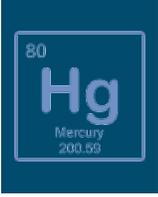
The table below summarizes the waste categorizations of the specific products available this season. Applying EPA's federal regulations to the flu vaccines, the vaccines meet the toxicity characteristic when the concentration of mercury is equal to or greater than 0.2mg/liter as a result of performing a test known as the toxicity characteristic leaching procedure (TCLP).

While we do not have results for such tests for vaccines, a calculation of the concentration of mercury in vaccines in which thimerosal is present as a preservative would cause the waste vaccine to fail the TCLP. Unless a manufacturer provides actual TCLP data indicating the mercury level to be below 0.2mg/liter or a healthcare facility has the test performed and the waste does not fail the TCLP, any vaccine waste containing thimerosal as a preservative should be managed as a toxic hazardous waste. As a result, all full or partially used multi-dose vials of the seasonal flu vaccine should be disposed of as federally hazardous waste, waste code D009 for mercury.



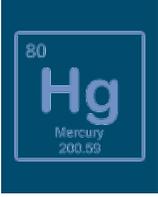
# Susceptible Populations

Population	Susceptibility Factors
Fetus / Developing Infant	Rapid neurodevelopment, immature blood-brain barrier, low glutathione, high brain accumulation
Pregnant Women	Placental transfer, fetal susceptibility, breast milk transfer
Young Children	Higher exposure per kg, immature detoxification systems
Older Adults	Reduced renal clearance, pre-existing neurodegenerative conditions
Genetically Susceptible	Impaired detox (e.g., GST, MTHFR, APOE4), mitochondrial dysfunction
High Fish Consumers	Bioaccumulation through predatory fish, dietary exposure
Occupationally Exposed Workers	Inhalation exposure, chronic accumulation (e.g., dentists, miners)



# Summary (1)

- Mercury is the third most toxic element on earth, behind polonium and plutonium, and has no physiological role in the human body.
- Thimerosal was grandfathered for use without adequate safety testing by the FDA.
- Thimerosal is not “generally recognized as safe or effective” (GRASE) by the FDA OTC Division since 1998.
- There is evidence that thimerosal is not an effective preservative at vaccine levels.
- Thimerosal can cross the placenta and blood brain barriers and converts to inorganic mercury in the brain at higher levels than methyl mercury.
- Studies have identified infants with blood levels after exposure to thimerosal that breach CDC guidelines for a case of mercury chemical poisoning.



## Summary (2)

- Thimerosal is recognized as a developmental and reproductive toxicant and is listed as a chemical on the California Proposition 65 list since 1990.
- Unused doses of thimerosal preserved flu shots must be disposed of as hazardous waste.
- Tremendous progress has been made in removing thimerosal, but more than 60,000 pregnant mothers receiving Medicaid in the 2019-2020 flu season received TCVs.
- We currently have enough thimerosal free flu vaccines to recommend that all pregnant women, infants and children receive only thimerosal free vaccines.
- After a critical appraisal of this issue almost 25 years ago, the prestigious Institute of Medicine made this same recommendation.
- Removing a known neurotoxin from being injected into our most vulnerable populations is a good place to start with Making America Healthy Again.