

CONGRESSIONAL TESTIMONY

U.S. HOUSE OF REPRESENTATIVES  
THE SUBCOMMITTEE ON LABOR, HHS, EDUCATION & RELATED  
AGENCIES  
COMMITTEE ON APPROPRIATIONS

OCTOBER 5, 2004 HEARING

*INFLUENZA VACCINE & ITS DELIVERY TO CHILDREN*

submitted by:

Mady Hornig, MD  
Director of Translational Research  
Jerome L. and Dawn Greene Infectious Disease Laboratory

Associate Professor of Epidemiology  
Mailman School of Public Health  
Columbia University

Chairman Regula, Representative Weldon, Representative Lowey, and Members of the Subcommittee, thank you for this opportunity to present information regarding our new animal model of the toxicity of the ethylmercury vaccine preservative, thimerosal, and its implications for human health. Our work addresses whether genes are important in determining if mercury exposures akin to those in childhood immunizations can disrupt brain development and function. I also submit for the record an electronic copy of the first paper published on this animal model in the Nature Publishing Group journal, *Molecular Psychiatry* (Hornig M, Chian D, Lipkin WI. Neurotoxic effects of postnatal thimerosal are mouse strain dependent. *Mol Psychiatry* 2004;9:833-845).

The premise of our research is that if mercury in vaccines creates risk for neurodevelopmental disorders such as autism, genetic differences are likely to contribute to that risk. We built upon an extensive, existing literature on toxicity of other forms of mercury in inbred mouse strains that affirmed the importance of specific genes controlling immune responses (major histocompatibility complex, or MHC) in determining mercury-induced autoimmune outcomes in mice. Earlier studies, however, did not use the form of mercury present in vaccines, known as thimerosal, and did not consider whether intramuscular, repetitive administration during early postnatal development, when the brain and immune systems are still maturing, might intensify toxicity. Based on reports of immune disturbances and family history of autoimmune disease in a subset of children with autism, we hypothesized that immune response genes linked to mercury immunotoxicity in mice would predict damage following low-dose, vaccine-based mercury in our mouse model.

Our predictions were confirmed. Using thimerosal dosages and timing that approximated the childhood immunization schedule, our model of postnatal thimerosal neurotoxicity demonstrated that the genes in mice that predict mercury-related immunotoxicity also predicted neurodevelopmental damage. Features reminiscent of those observed in autism occurred in the mice of the genetically sensitive strain, including: generalized behavioral impoverishment and abnormal reaction to novel environments; enlargement of the hippocampus, a region of the brain involved in learning and memory; correlation of hippocampal enlargement with abnormalities in exploration and anxiety; increased packing density of neurons in hippocampus; and disturbances in glutamate receptors and transporters. Only mice carrying the H-2<sup>s</sup> susceptibility gene showed these autism-like effects (SJL/J mice). Two mouse strains with different H-2 genes (C57BL6/J mice, H-2<sup>b</sup>; BALB/cJ mice, H-2<sup>d</sup>) did not demonstrate adverse consequences following thimerosal exposure.

It is important to emphasize that these animal model studies do not provide conclusive evidence regarding a link between mercury exposure and human autism. Nonetheless, the finding that a specific genetic constraint profoundly alters the brains and behavior of thimerosal-exposed mice confirms the biological plausibility of thimerosal neurotoxicity, provides critical guidance for the interpretation of existing epidemiologic investigations into the potential association of thimerosal with neurodevelopmental disorders, and suggests important new avenues for future research. Our work implies that if genetic factors are operative in mediating a link between thimerosal and autism in humans, then epidemiologic studies that fail to consider genetic susceptibility factors will be compromised in their ability to detect a statistically significant effect even if one exists.

In this context, it is worthwhile to recall the example of acrodynia or “pink disease,” a syndrome described in 1 of 500 infants exposed to mercury (Hg<sub>2</sub>Cl<sub>2</sub>, calomel) in teething powders in the early 1900s. Acrodynia disappeared with the removal of such products from the market. Genetic factors are suspected as determinants of susceptibility.

Recent findings, presented at scientific meetings but as yet unpublished, suggest that thimerosal neurotoxicity in susceptible mice involves the generation of autoantibodies targeting brain components. This autoimmune response persists long after the presence of mercury can no longer be detected. If confirmed, these findings will enable us to develop a human diagnostic test to determine whether some individuals with autism have similar autoantibodies present in their peripheral blood. Such work would not only bring us a step closer to identifying the genes associated with thimerosal neurotoxicity in humans, facilitating prevention programs, it would also validate the utility of this animal model for the development of safe and effective modes of intervention.

It is highly likely that the neurotoxic effects of cumulative mercury burden, including exposure to other sources or forms of mercury (thimerosal in products other than vaccines; methylmercury in contaminated fish), follow similar patterns of genetic restriction; it is also likely that similar genetic factors influence the neurotoxicity observed following exposure to xenobiotics other than mercury (e.g., PCBs, the PBDEs used as flame retardants in computers, and, possibly, infectious agents). Age and developmental status at the time of exposure, nutritional factors, and gender are also known to influence outcomes. We have limited ability to explain the interplay of such factors in humans; consider the example of the disparate cognitive outcomes reported in children in the Faeroe Islands and the Seychelles after similar prenatal methylmercury exposures. The reasons for this divergence remain unclear. The design of future epidemiologic studies must take into account the possibility of multiple xenobiotic exposures as well as the influence of factors that modulate risk. Our studies have important implications for understanding the role of gene-environment interactions in the pathogenesis of autism and related neurodevelopmental disorders, and for ensuring that our vaccines meet the highest standards of safety.

I refer Subcommittee Members to our recent publication in *Molecular Psychiatry* where experimental findings and their implications are discussed in more detail.

Thank you for your attention.  
Mady Hornig, MD  
New York, NY