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IMMEDIATE COMMUNICATION

Neurotoxic effects of postnatal thimerosal are mouse strain dependent

M Hornig[1], D Chian[1] and WI Lipkin[1,2]

[1]*Jerome L and Dawn Greene Infectious Disease Laboratory, Department of Epidemiology, Mailman School of Public Health, Columbia University, New York, NY, USA;* [2]*Departments of Neurology and Pathology, Columbia University College of Physicians and Surgeons, New York, NY, USA*

The developing brain is uniquely susceptible to the neurotoxic hazard posed by mercurials. Host differences in maturation, metabolism, nutrition, sex, and autoimmunity influence outcomes. How population-based variability affects the safety of the ethylmercury-containing vaccine preservative, thimerosal, is unknown. Reported increases in the prevalence of autism, a highly heritable neuropsychiatric condition, are intensifying public focus on environmental exposures such as thimerosal. Immune profiles and family history in autism are frequently consistent with autoimmunity. We hypothesized that autoimmune propensity influences outcomes in mice following thimerosal challenges that mimic routine childhood immunizations. Autoimmune disease-sensitive SJL/J mice showed growth delay; reduced locomotion; exaggerated response to novelty; and densely packed, hyperchromic hippocampal neurons with altered glutamate receptors and transporters. Strains resistant to autoimmunity, C57BL/6J and BALB/cJ, were not susceptible. These findings implicate genetic influences and provide a model for investigating thimerosal-related neurotoxicity.
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Autism spectrum disorders (ASDs) comprise a set of highly heritable conditions[1] with core impairments in social interaction, communication, and imagination. The prevalence of ASDs is reported to be rising worldwide,[2–4] an increase not fully explained by changes in awareness and diagnostic patterns.[2,3,5,6] Environmental susceptibility genes may be determinants of adverse neurodevelopmental outcomes following pre- or postnatal exposures. One environmental factor may be increased mercury burden through industrial sources, fish, and sodium ethylmercurithiosalicylate (thimerosal), a preservative recently eliminated from many vaccines.[7,8] A large, retrospective study investigating the association of thimerosal-containing vaccines and neurodevelopmental outcomes in an unselected cohort was inconclusive and called for further research into vulnerability factors.[9] An autoimmune diathesis is described in ASD probands[10–15] and their first degree relatives.[16,17] Major histocompatibility complex (MHC) genes regulate risk of mercury-induced autoimmunity in mice.[18,19] To examine whether immunogenetic factors mediate vulnerability to mercury-related neurodevelopmental damage, we exposed mice of differing MHC (H-2) backgrounds[20] to thimerosal in doses and timing equivalent to the pediatric immunization schedule. Profound behavioral and neuropathologic disturbances were observed after postnatal thimerosal in SJL/J (H-2^s) mice, but not in strains without autoimmune sensitivity (BALB/cJ, H-2^d, or C57BL/6J, H-2^b mice).

Correspondence: M Hornig, Jerome L and Dawn Greene Infectious Disease Laboratory, Department of Epidemiology, Mailman School of Public Health, Columbia University, 722 West 168th Street, 18th Floor, New York, NY 10032 USA.
E-mail: mady.hornig@columbia.edu
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Materials and methods

Animals

Mouse pups of SJL/J (SJL), C57BL6/J (C57), and BALB/cJ (BALB) strains (Jackson Labs) were inoculated intramuscularly (i.m.) at postnatal day (P)7, P9, P11, and P15 with: (1) *thimerosal-only* (*Thim-only*), 14.2, 10.8, 9.2, or 5.6 μg/kg of ethylmercury per postnatal immunization day, respectively, as sodium ethylmercurithiosalicylate (Sigma); (2) *thimerosal-vaccine* (*Thim-vax*), thimerosal-preserved Diphtheria, Tetanus, acellular Pertussis (DTaP, Lederle), and Haemophilus influenza B (HiB, Lederle) vaccines; or (3) *Control* (phosphate-buffered saline (PBS)) in 25–50 μl volume. Administration days were based on the 2001 US immunization schedule:[7] two, four, six, and 12 months of age. Differences between human and murine central nervous[21] and immune system[22] maturation, including neuronal migration and window of immune tolerance, guided time point selection. P7 in mice is expected to be after most