Safety of Thimerosal-Containing Vaccines: A Two-Phased Study of Computerized Health Maintenance Organization Databases

Thomas Verstraeten, MD*‡; Robert L. Davis, MD, MPH§; Frank DeStefano, MD, MPH‡; Tracy A. Lieu, MD, MPH‖; Philip H. Rhodes, PhD‡; Steven B. Black, MD¶; Henry Shinefield, MD¶; and Robert T. Chen, MD‡; for the Vaccine Safety Datalink Team

ABSTRACT. *Objective.* **To assess the possible toxicity of thimerosal-containing vaccines (TCVs) among infants.**

Methods. **A 2-phased retrospective cohort study was conducted using computerized health maintenance organization (HMO) databases. Phase I screened for associations between neurodevelopmental disorders and thimerosal exposure among 124 170 infants who were born during 1992 to 1999 at 2 HMOs (A and B). In phase II, the most common disorders associated with exposure in phase I were reevaluated among 16 717 children who were born during 1991 to 1997 in another HMO (C). Relative risks for neurodevelopmental disorders were calculated per increase of 12.5 μg of estimated cumulative mercury exposure from TCVs in the first, third, and seventh months of life.**

Results. **In phase I at HMO A, cumulative exposure at 3 months resulted in a significant positive association with tics (relative risk [RR]: 1.89; 95% confidence interval [CI]: 1.05–3.38). At HMO B, increased risks of language delay were found for cumulative exposure at 3 months (RR: 1.13; 95% CI: 1.01–1.27) and 7 months (RR: 1.07; 95% CI: 1.01–1.13). In phase II at HMO C, no significant associations were found. In no analyses were significant increased risks found for autism or attention-deficit disorder.**

Conclusions. **No consistent significant associations were found between TCVs and neurodevelopmental outcomes. Conflicting results were found at different HMOs for certain outcomes. For resolving the conflicting findings, studies with uniform neurodevelopmental assessments of children with a range of cumulative thimerosal exposures are needed.** ***Pediatrics*** **2003;112:1039–1048;** ***cohort study, computerized medical record systems, language development disorders, speech disorders, thimerosal, vaccines.***

ABBREVIATIONS. Hg, mercury; EPA, Environmental Protection Agency; TCV, thimerosal-containing vaccine; HMO, health maintenance organization; VSD, Vaccine Safety Datalink; CDC, Centers for Disease Control and Prevention; LBW, low birth weight; ICD-9-CM, *International Classification of Diseases, Ninth Revision, Clinical Modification*; ADD, attention-deficit disorder; RR, relative risk; HBV, hepatitis B vaccine.

From the *Epidemic Intelligence Service Program, Epidemiology Program Office, Centers for Disease Control and Prevention, Atlanta, Georgia; ‡Vaccine Safety and Development Activity, Epidemiology and Surveillance Division, National Immunization Program, Centers for Disease Control and Prevention, Atlanta, Georgia; §University of Washington and Group Health Cooperative of Puget Sound, Seattle, Washington; ‖Center for Child Health Care Studies, Department of Ambulatory Care and Prevention, Harvard Pilgrim Health Care and Harvard Medical School, and Division of General Pediatrics, Children's Hospital, Boston, Massachusetts; and ¶Kaiser Permanente Vaccine Study Center, Oakland, California.

Received for publication Jan 24, 2003; accepted Jul 9, 2003.

Reprint requests to (F.D.) Centers for Disease Control and Prevention, 1600 Clifton Rd, NE, MS E-61, Atlanta, GA 30333. E-mail: fxd1@cdc.gov

From the 1930s to the late 1990s, many routinely administered vaccines in the United States contained thimerosal, an organic compound that is 49% mercury (Hg) by weight and is metabolized to ethylmercury and thiosalicylate. To meet Food and Drug Administration guidelines, thimerosal was added to vaccines to prevent bacterial and fungal contamination of multidose vaccine vials (except live viral vaccines).[1] Another organic Hg compound, methylmercury, has been found in studies of fish and grain ingestion to affect human neurologic and renal systems.[2–4] These studies, along with studies of prenatal Hg exposure, have been used by regulatory agencies to develop guidelines on exposure limits for methylmercury, the most stringent of which was set by the Environmental Protection Agency (EPA).[5–8] During a Food and Drug Administration review of Hg and other metals in drugs, it was determined that some infant immunization schedules that use thimerosal-containing vaccines (TCVs) adopted in 1991 may have exceeded the 1995 EPA guidelines for exposure to organic Hg (1 μg/kg/d vs 3 μg/kg/d in the previous 1985 EPA guidelines).[1,9–11] In July 1999, the American Academy of Pediatrics and the US Public Health Service recommended removing thimerosal from childhood vaccines as soon as possible as a precautionary measure.[12–14]

Although oral ingestion of organic Hg has been studied, information concerning the effects of parenteral exposure to these compounds in humans is limited to a few case reports,[15–18] none of which involved exposure from vaccines. Vaccines, however, constitute a nearly universal exposure for children in the United States and most other countries. To evaluate the theoretical concerns of the potentially toxic effects of thimerosal in vaccines, we studied neurodevelopmental outcomes among a large group of children with documented exposure to varying levels of thimerosal from vaccinations in several health maintenance organizations (HMOs).

METHODS

The study was conducted in 2 phases. In phase I, a number of neurodevelopmental disorders were identified a priori as possibly related to ethylmercury exposure.[2–5,7] In this phase, using primar-

ily preexisting HMO administrative databases collected for the Vaccine Safety Datalink (VSD) project, we screened for potential associations between these disorders and cumulative thimerosal exposure by 1, 3, and 7 months of age.[10] In the second phase, we attempted to confirm selected positive associations seen in phase l between thimerosal exposure and these outcomes in another independent cohort of HMO children (phase II) with similar largely preexisting HMO administrative data. Because of the smaller size of this second cohort, we were able to evaluate only the most common outcomes associated with thimerosal in phase I.

Study Participants

For phase I, we studied a cohort of infants from the VSD project, which was created in 1991 by the National Immunization Program of the Centers for Disease Control and Prevention (CDC). The VSD methods have been described previously.[19–21] The project links medical event information, specific vaccine history (including manufacturer and lot number), and selected demographic information from the computerized databases of several HMOs. Because most of the neurodevelopmental outcomes of interest would have been cared for only in the outpatient setting, we restricted our analyses to children who were born from January 1992 through December 1998 at the 2 HMOs (HMO A and HMO B) with the most complete computerized outpatient data. At HMO A, clinic data for outcomes were available throughout the study period; for HMO B, clinic data were available starting in January 1995. For both HMOs, children had follow-up data through the end of 2000. For phase II, we used computer databases similar to those of the VSD to study children in a third HMO (HMO C), where data were available on children who were born from January 1991 through December 1997, with follow-up through May 1998.

To capture all vaccinations in the first year of life, we restricted the cohorts to children who were born into the HMO and remained enrolled continuously for the first year of life. To be certain that we studied children who actually received most of their primary care through the HMO, we excluded children who did not have documentation in the HMO databases of at least 2 polio vaccines by the age of 1 year.

We excluded from the main analysis infants with low birth weight (LBW) of <2500 g and those with a diagnosis of a congenital or severe perinatal disorder or born to mothers with serious medical problems of pregnancy (Appendix 1). We performed a separate analysis of infants with birth weights between 1500 and 2499 g.

Exposure Assessment

We assessed cumulative exposure at 1, 3, and 7 months of life, when the exposure burden relative to body weight was highest. During the years of the study, the HMOs routinely used multidose vials for the vaccines of interest, and the exposure estimates were based on the mean Hg content of each vaccine in multidose vials (Table 1).

Outcome Assessment

We identified the *International Classification of Diseases, Ninth Revision, Clinical Modification* (ICD-9-CM) diagnosis codes in the VSD database for HMOs A and B and the Costar codes in HMO C's database that were associated with the a priori selected neurodevelopmental disorders. The automated administrative databases that were used for this study included diagnoses made in the clinic, emergency department, and hospital.[19,20] Because a diagnosis of most of these conditions in the first year of life may be less reliable than later diagnoses, we included only diagnoses that were made past the age of 12 months.

Speech and language disorders were important outcomes, but coding practices for these conditions varied by HMO. At HMO B, separate codes for language delay (ICD-9 315.31) and speech delay (315.39) were used. At HMO A, only the code for speech delay was used, and there were no language delay codes. At HMO C, the Costar code was for combined language and speech disorders. Theoretically, the "language delay" code should be indicative of problems with expressive language development (eg, vocabulary, tense, word recall, sentence length and complexity) and the "speech delay" code should indicate difficulties or delays in development of speech sounds appropriate for age (eg, substituting one sound for another, omission of final consonants). The distinction between the diagnostic terms attention-deficit/hyperactivity disorder and attention-deficit disorder (ADD) can also be confusing. In this report, we use the term ADD to be consistent with the ICD-9 code (314.0) that we used in our analyses.

To assess the validity of the computerized diagnoses, we reviewed medical charts for selected diagnoses codes ascertained through 1998. For speech and language delay, autism, and ADD, we reviewed the medical charts of all 618 children in HMOs A and B and 826 children in HMO C with at least 2 automated diagnoses of speech delay, and a sample of 377 children in HMOs A and B and 100 children in HMO C with at least 1 automated diagnosis of ADD, and 120 children in HMOs A and B with at least 1 automated diagnosis of autism. For verification, we required documentation in the medical record that the diagnosis was made by an appropriate clinical or behavioral specialist.

Statistical Analyses

In the primary analyses, relative risks (RRs) were calculated for the cumulative exposure to thimerosal by 1, 3, and 7 months of age. Because of power considerations, we decided a priori to perform an evaluation only of the cumulative effect of thimerosal exposure on the risk of outcomes with at least 50 or more cases. We estimated RRs separately for each HMO, using proportional hazards models stratified by sex and year and month of birth at HMO A and by sex and year and month of birth and clinic most

TABLE 1. Hg Exposure From TCVs for Children Following the Recommended Immunization Schedule in the First 7 Months of Life, HMOs A and B, 1992–1999

Age at Exposure	Vaccines (Dose)	Total Hg Dose in the Period	Cumulative Hg Dose at End of the Period
First mo	HBV (First dose)	12.5 μg	12.5 μg
2–3 mo	DTP and Hib (first dose) HBV (second dose)*	25, 37.5, 50, or 62.5 μg†	37.5, 50, 62.5, or 75 μg†
4–5 mo	DTP and Hib (second dose) HBV (second dose)*	25, 37.5, 50, or 62.5 μg‡	75 or 125 μg‡
6–7 mo	DTP (third dose) Hib (third dose) HBV (third dose)	25, 50, or 62.5 μg§	112.5 or 187.5 μg§

DTP indicates combined diphtheria, tetanus, and Pertussis vaccine; Hib, *Haemophilus influenzae* type B vaccine.

* HBV second dose can be administered between months 1 and 4.

† Depending on whether DTP and Hib were given separately (both contain 25 μg of Hg) or as a combination vaccine (containing 25 μg of Hg) and the timing of the second HBV dose.

‡ Depending on whether DTP and Hib were given separately or as a combination vaccine and assuming that all 3 doses are given as combined or separate.

§ Depending on whether DTP and Hib were given separately or as a combination vaccine and assuming that all 3 doses are given as combined or separate and whether the third HBV dose was given before 7 months.

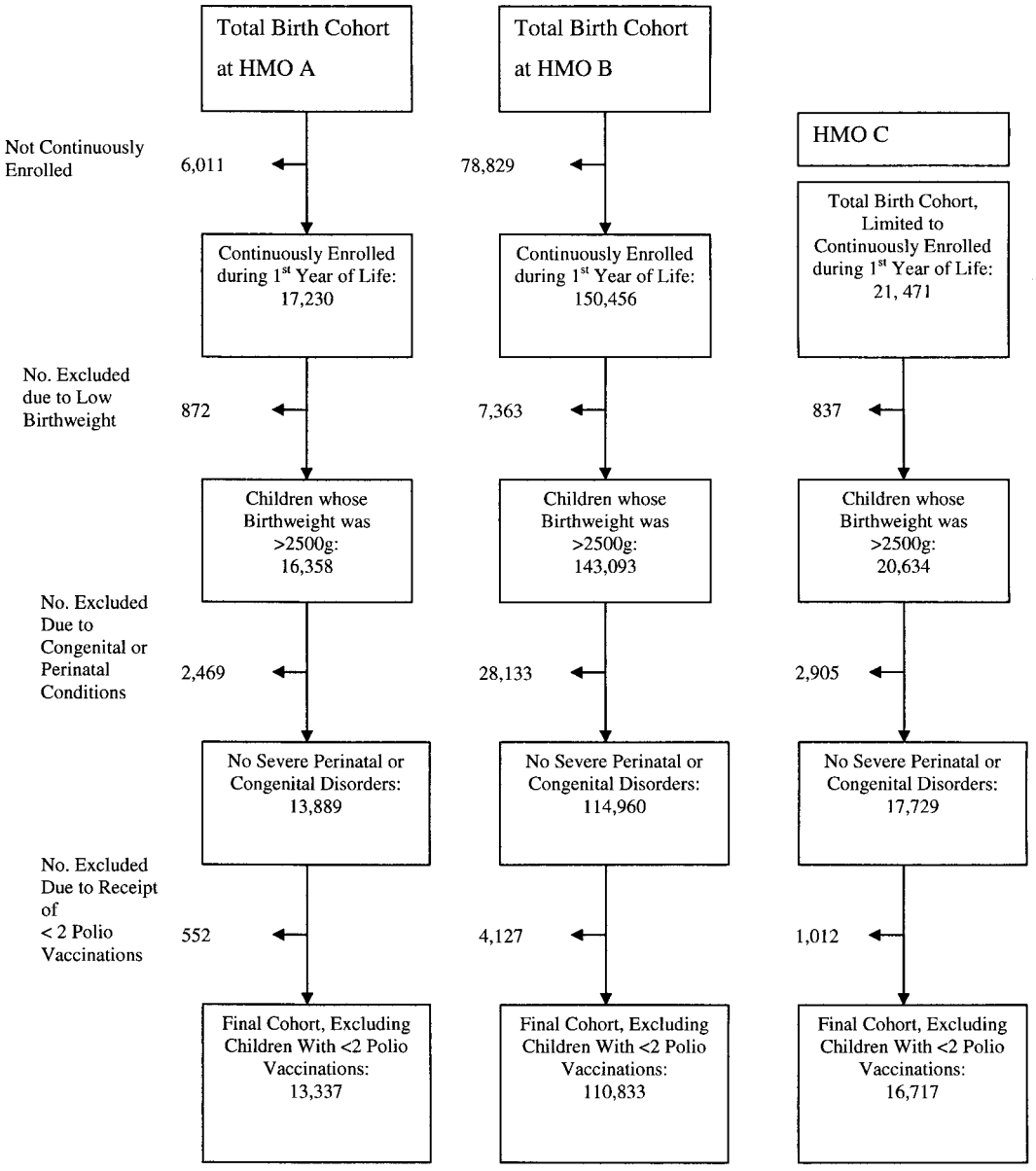

Fig 1. Creation of study cohorts at HMOs A, B, and C.

often visited at HMO B. The time variable in the models started at the first birthday for children in HMO A or at the first birthday or January 1, 1995 (whichever came later), for children at HMO B; for children in each HMO, the time of follow-up ended at the date of diagnosis or the last date of follow-up. Temporary disenrollment with reenrollment was allowed, but person-time and diagnoses were used only while the child was enrolled in the HMO. We used $P < .05$ to define statistical significance.

We were concerned that parents who had their children vaccinated on time (and therefore were more likely to have increased thimerosal exposure at each of the time periods studied) were also more likely to seek medical care for common pediatric ambulatory conditions. Support for this concern was provided by analyses indicating that in each year from 1994 to 1998, children who received >75 μg Hg in the first 7 months of life, compared with children who received 0 to 75 μg Hg in the first 7 months of life, had significantly more well child care visits and significantly more visits for "upper respiratory infections" in both the second and third years of life (Appendix 2). To try to control for health care-seeking behavior, we performed the analyses in phase I restricted to children who had made at least 1 visit to a clinic or an emergency department at the same month of age as cases. For phase II, this extent of health care visits data was not available in the analytic data set and no such adjustment for health care-seeking behavior was possible. We also were not able to make such adjustments in the subanalysis of LBW infants.

To simplify the presentation of the results given the large number of outcomes studied and the different exposure time periods that were assessed, we modeled exposure as a continuous variable with increments of 12.5 μg Hg. To illustrate the change in risk with each level of exposure and as a visual check of the linearity assumption made in analyses of exposure as a continuous variable, we also performed additional analyses in which we modeled exposure as a categorical variable. For these analyses, the exposure levels were 0 to 25 μg, 37.5 to 50 μg, and ≥62.5 μg at 3 months and 0 to 75 μg, 87 to 162.5 μg, and ≥175 μg at 7 months, respectively. We restricted these analyses to outcomes for which significant associations were found in the analysis of exposure as a linear variable and certain outcomes of particular interest (eg, ADD, autism).

In the analyses restricted to moderately LBW infants, we included children who weighed from 1500 to 2499 g at birth, were enrolled in the HMO in the first month of life and remained enrolled past 1 year of age, and had 2 or more polio vaccinations by 1 year. We were not able to maintain the other exclusion criteria listed in Appendix 1 because insufficient numbers would have remained in the analysis. The statistical analyses were stratified by

TABLE 2. Number of Children, Age at Diagnosis, and Proportion of Boys Diagnosed at HMOs A (1992–2000) and B (1995–2000)

ICD-9 Code	Outcome	HMO A			HMO B		
		N	Age*	% Boys	*N*	Age*	% Boys
All children		13 337		51	110 833		50
299.0	Autism	21	49	90	202	44	80
299.8	Other childhood psychosis	20	54	70	108	55	94
307.0	Stammering	61	40	70	112	39	69
307.2	Tics	62	61	60	201	62	75
307.4	Sleep disorders	70	32	56	159	29	58
307.5	Eating disorders	20	33	30	82	26	54
313.8	Emotional disturbances	84	67	73	320	64	78
314.0	ADD	170	72	78	940	70	79
315.31	Developmental language delay	35	62	60	586	32	75
315.39	Developmental speech delay	694	38	68	1941	31	72
315.3	Speech or language delay	730	38	68	2288	31	72
315.4	Coordination disorder	83	35	64	26	56	73

* Median age at first diagnosis, in months.

HMO, year of birth, and sex and controlled for birth weight (250-g intervals).

RESULTS

Phase I: HMOs A and B

Cohort Selection

A total of 252 526 children (23 241 at HMO A and 229 285 at HMO B) were born into the 2 VSD HMOs (Fig 1). After all exclusion criteria were applied, the final study cohort size was 13 337 at HMO A and 110 833 at HMO B.

Outcome Assessment

At HMO A, 8 categories of neurodevelopmental disorders contained 50 or more children; in HMO B, there were 11 such categories (Table 2). The most frequent diagnoses were those of speech delay followed by ADD. As noted previously, there were substantive differences in the proportions of children at HMOs A and B who had a diagnosis of speech delay or of language delay. The median age at first diagnosis for the children within the study cohorts varied from 26 months for eating disorders to 72 months for ADD. For each category of neurodevelopmental disorders (with the exception of one), more boys than girls received a diagnosis of neurodevelopmental disorders. For the children whose charts were reviewed, the confirmation rates for speech delay, autism, and ADD were 81.6%, 92.3%, and 42.1% for HMO A and 66.8%, 81.3%, and 28.2% for HMO B, respectively.

Risk Estimates

Tables 3 and 4 show the adjusted RRs associated with cumulative thimerosal exposure by 1, 3, and 7 months of age. At HMO A, a significantly increased risk was seen only with cumulative exposure at 3 months and the diagnosis of tics. At HMO B, significantly increased risks were seen with cumulative exposure at 3 and 7 months and language delay.

In the categorical analyses of cumulative exposure at 3 months of age at HMO B (Table 5), there was a significant association between the highest level of exposure (≥62.5 micrograms) and language delay. For the categorical analyses of cumulative exposure at 7 months of age (Table 5), there was a borderline statistically significant negative association of speech delay with medium and high levels of thimerosal exposure at HMO A. There were no significant associations between exposure and ADD.

There were sufficient cases for analysis of autism only at HMO B. No significant associations were found with cumulative exposure at any age and risk for autism in either the continuous (Table 4) or the categorical analyses (Table 5).

TABLE 3. RRs by Increase of 12.5 μg of Hg Exposure From TCVs at HMO A

Outcome	1-Month Cumulative Hg		3-Month Cumulative Hg		7-Month Cumulative Hg	
	RR	95% CI	RR	95% CI	RR	95% CI
Stammering	0.89	0.40–1.97	1.18	0.74–1.89	1.17	0.97–1.41
Tics	1.25	0.47–3.29	1.89*	1.05–3.38	1.12	0.93–1.34
Sleep disorders	0.79	0.38–1.61	0.93	0.71–1.21	1.08	0.95–1.24
Emotional disturbances	1.00	0.42–2.36	0.98	0.66–1.45	0.92	0.81–1.03
ADD	0.92	0.52–1.59	0.83	0.68–1.02	0.93	0.84–1.02
Speech delay	1.07	0.83–1.38	1.03	0.93–1.15	0.97	0.92–1.01
Speech/language delay	1.14	0.88–1.46	1.03	0.93–1.14	0.97	0.93–1.02
Coordination disorders	1.67	0.78–3.57	1.19	0.82–1.71	1.00	0.87–1.15

CI indicates confidence interval.
* $P < .05$.

TABLE 4. RRs by Increase of 12.5 μg of Hg Exposure From TCVs at HMO B

Outcome	1-Month Cumulative Hg		3-Month Cumulative Hg		7-Month Cumulative Hg	
	RR	95% CI	RR	95% CI	RR	95% CI
Autism	1.16	0.78–1.71	1.06	0.88–1.28	1.00	0.90–1.09
Other child psychosis	1.03	0.60–1.74	0.93	0.73–1.19	1.04	0.91–1.20
Stammering	0.61	0.33–1.14	1.10	0.86–1.41	1.06	0.93–1.21
Tics	0.85	0.55–1.30	0.95	0.78–1.15	1.09	0.98–1.21
Sleep disorders	1.24	0.80–1.93	1.15	0.95–1.39	1.09	0.99–1.19
Eating disorders	0.90	0.50–1.61	0.97	0.72–1.29	0.98	0.85–1.14
Emotional disturbances	0.76	0.54–1.07	1.02	0.88–1.18	1.01	0.93–1.10
ADD	0.90	0.74–1.10	1.01	0.93–1.11	1.02	0.97–1.07
Language delay	1.06	0.83–1.35	1.13*	1.01–1.27	1.07*	1.01–1.13
Speech delay	1.02	0.90–1.17	1.04	0.98–1.10	1.02	0.99–1.05
Language/speech delay	1.03	0.91–1.17	1.05	0.99–1.11	1.02	0.99–1.05

* $P < .05$.

TABLE 5. RRs by Category of Cumulative Hg Exposure at 3 and 7 Months

Outcome	Hg (μg)	HMO A				HMO B			
		RR 95% CI	*N*	χ^2	*P* Value	RR 95% CI	*N*	χ^2	*P* Value
3 Months									
	0–25	1.00	61	0.13	.94	1.00	106	2.28	.32
Speech delay	37.5–50	1.04 (0.61–1.75)	107			1.14 (0.91–1.44)	1297		
	≥62.5	1.09 (0.65–1.81)	526			1.21 (0.93–1.58)	538		
	0–25		1*			1.00	24	5.44	.07
Language delay	37.5–50		6			1.44 (0.90–2.28)	419		
	≥62.5		28			1.87 (1.08–3.23)	143		
ADD	0–25	1.00	5	4.36	.11	1.00	55	0.33	.85
	37.5–50	0.88 (0.27–2.79)	60			1.00 (0.71–1.39)	627		
	≥62.5	0.41 (0.13–1.20)	105			1.08 (0.72–1.61)	258		
	0–25		1			1.00	11	1.84	.40
Autism	37.5–50		5			1.61 (0.77–3.34)	158		
	≥62.5		15			1.38 (0.55–3.48)	33		
7 Months									
	0–75	1.00	68	5.37	.07	1.00	250	2.23	.33
Speech Delay	87–162.5	0.58 (0.37–0.93)	202			1.11 (0.95–1.30)	1362		
	≥175	0.58 (0.36–0.92)	424			1.04 (0.82–1.32)	329		
Language delay	0–75		1*			1.00	75	2.34	.31
	87–162.5		10			1.20 (0.91–1.59)	422		
	≥175		24			1.37 (0.87–2.14)	89		
ADD	0–75	1.00	2	2.33	.31	1.00	101	2.51	.28
	87–162.5	1.64 (0.32–8.28)	82			1.22 (0.95–1.57)	684		
	≥175	1.19 (0.23–6.05)	86			1.21 (0.83–1.76)	155		
	0–75		1			1.00	37	1.08	.58
Autism	87–162.5		8			0.95 (0.62–1.46)	148		
	≥175		12			0.65 (0.27–1.52)	17		

* No comparisons were made when fewer than 50 children received a diagnosis of a condition.

Phase II: HMO C

Cohort Selection

A total of 21 471 children were born into HMO C and were also continuously enrolled for the first year of life (the numbers of the entire birth cohort, including those not continuously enrolled for the first year of life, were unavailable). After children who did not receive at least 2 polio vaccinations in the first year or who had LBW or a serious congenital or perinatal anomaly were excluded, the final study cohort size was 16 717 (Fig 1).

Outcome Assessment

A total of 1134 children had a speech/language delay, 91 children had stammering, 499 children had sleep disorders, and 97 children had ADD from the computerized clinic records. The median age at first diagnosis was 24 months for speech or language disorder, 50 months for ADD, 35 months for stammering, and 19 months for sleep disorder, and similar to phase 1, there was a male excess for each disorder. Among the children for whom medical records were reviewed, we confirmed 647 (78%) and 44 (44%) of the automated diagnoses of speech or language delay and ADD, respectively.

Risk Estimates

There were no significant associations between cumulative thimerosal exposures at 1, 3, or 7 months of age and speech/language disorder, ADD, stammering, or sleep disorder (Table 6).

Analysis of LBW Infants: HMOs A, B, and C

In the subanalysis of LBW infants, a limited number of outcomes could be evaluated because of sample size constraints. Restricting the analyses to conditions with at least 50 cases, we evaluated risk for cumulative mercury exposure at 3 and 7 months by

TABLE 6. RRs by Increase of 12.5 μg of Hg Exposure From TCVs at HMO C

Outcome	1-Month Cumulative Hg		3-Months Cumulative Hg		7-Months Cumulative Hg	
	RR	95% CI	RR	95% CI	RR	95% CI
Stammering	0.77	0.47–1.26	0.97	0.78–1.20	0.99	0.88–1.10
Tics	0.93	0.45–1.92	1.26	0.81–1.94	1.18	0.97–1.42
Sleep disorders	0.97	0.79–1.19	1.02	0.92–1.13	1.05	0.99–1.11
ADD	0.88	0.53–1.48	0.96	0.79–1.18	0.96	0.87–1.05
Speech/language delay	0.91	0.79–1.04	0.96	0.90–1.02	0.98	0.94–1.01

12.5-μg increase (Table 7). We were able to evaluate the combined outcome of speech or language delay at all 3 HMOs and ADD at HMO B. We found no statistically significant increased risks for either outcome.

DISCUSSION

In this analysis using computerized HMO databases to screen for possible associations between exposure to thimerosal in infant vaccines and neurodevelopmental outcomes, we did not find evidence of a clear association between thimerosal and specific neurodevelopmental disorders. In the first phase of our study, we observed an association between thimerosal exposure and some of the neurodevelopmental disorders screened, most notably between cumulative thimerosal exposure by 3 and 7 months of age and speech and language disorders at 1 HMO, and also an association between cumulative thimerosal exposure by 3 months of age in 1 HMO and tic disorder. The results between HMOs, however, were inconsistent. Our study encompassed a large number of separate analyses and, by chance alone, at least some associations would be expected to be statistically significant. We did not adjust the level of statistical significance of our estimates for the multiple comparisons made but chose instead to attempt to confirm our positive findings in an independent third HMO. In the second phase of this study, no associations that had been seen previously in either of the first 2 HMOs were detected at the third HMO.

The discrepant findings have several possible explanations, including differences in outcome ascertainment. HMO B is the only HMO in our study where speech therapy is not covered by the health plan. Because such therapy is not provided, primary care providers in this HMO may have screened less aggressively for speech or language disorders among young children. Thus, parental concern may have been a more important factor in the ascertainment of these disorders. If parents at this HMO who were more concerned about subtle neurodevelopmental delays were also more likely to adhere to a timely vaccination schedule, then ascertainment bias might have resulted in falsely elevated estimates of the association between thimerosal and these disorders. We attempted to control for differences in health care-seeking behavior by matching on clinic visits. Nevertheless, some significant associations remained for language delay.

The biological plausibility of the small doses of ethylmercury present in vaccines leading to increased risks of neurodevelopmental disorders is uncertain. The effect of organic Hg on neurologic development has been the focus of several studies.[5,7,22–24] Two prospective cohort studies of prenatal exposure to methylmercury from fish consumption have resulted in conflicting findings. In the Seychelles, Davidson et al[5,25] found no effect of pre- or postnatal methylmercury exposure on the neurologic development of 711 children at 66 months of age. In the Faroe Islands, Grandjean and colleagues[7,9,26] found an adverse effect of prenatal exposure to methylmercury on attention, language, and memory at 7 years of age among 917 children. Attention was also found to be inversely related to hair Hg concentrations in Amazonian children aged 7 to 12 years,[23] and speech retardation by 24 months was related to maternal hair Hg concentrations in Iraqi children.[27]

All of these and other studies involved ingested methylmercury, and their relevance to our study of ethylmercury bolus exposure by injection of TCVs is unknown. The magnitudes of Hg exposure in these other studies were also much higher than Hg exposure from vaccines. For example, blood Hg levels

TABLE 7. RRs by Increase of 12.5 μg of Hg Exposure from TCVs for Selected Outcomes Among Moderately Low Birth Weight Infants (1500–2499 g)

Outcome	HMO	Cases (*n*)	3-Months Cumulative Hg		7-Months Cumulative Hg	
			RR	95% CI*	RR	95% CI
Speech or language delay	A	55	1.09	0.86–1.37	1.04	0.92–1.17
	B	194	0.93	0.82–1.06	0.98	0.91–1.05
	C	65	0.97	0.79–1.19	1.04	0.90–1.19
ADD	B	64	0.99	0.75–1.29	0.99	0.83–1.17

* RR (95% CI) from proportional hazards regression models stratified by year of birth and sex, and adjusted for birth weight (250-g increments). The ADD results were also stratified by usual clinic and controlled for health care–seeking behavior.

after hepatitis B vaccine (HBV) in newborns, as measured by Stajich and colleagues,[7,9,22,28,29] although significantly elevated, were far below "no effect" levels as determined by the studies in the Faroe and Seychelles Islands. The results of a recently published study suggest that ethylmercury from thimerosal is metabolized and cleared from children more rapidly than methylmercury.[30] The Immunization Safety Review Committee of the Institute of Medicine concluded that although the evidence is indirect and incomplete, the hypothesis that TCVs could be associated with neurodevelopmental disorders is biologically plausible.[31]

Our use of automated databases has a number of limitations. As most vaccines used in the study population were either thimerosal-free throughout our study period (eg, polio) or thimerosal containing throughout our study period (eg, multidose HBV), our main analyses did not differentiate between the effect of thimerosal and other vaccine components. For example, we did not differentiate potential effects of thimerosal from those of whole-cell pertussis vaccine, which has been associated with an increased risk of encephalopathy. Encephalopathy after pertussis vaccination, however, is rare and unlikely to have had a meaningful impact on our results.[32]

To try to isolate the effects of thimerosal from other vaccine constituents, we performed a subanalysis comparing risks associated with diphtheria-tetanus-whole cell pertussis vaccine or diphtheria-tetanus-acellular pertussis vaccine and *Haemophilus influenzae* type b vaccine given separately or combined (Appendix 3). The 2 vaccination regimens included the same vaccine antigens but differed by Hg content (25 μg for the combined vaccine vs a total of 50 μg when the 2 vaccines were given separately). Only at HMO B were both the combined and separate products used. In the analyses of speech and language delay and ADD with cumulative exposure by 3 months, we did not find any statistically significant increased risks associated with increase in Hg exposure when the 2 vaccines were given separately compared with combined.

We evaluated the effect of the study exclusion criteria to determine whether they had an undue influence on our study findings. For the outcomes of speech delay and/or language delay, there was no appreciable effect on the observed RR of any of the exclusion criteria (Appendix 4). A similar analysis for autism also found no appreciable effect of the exclusion criteria (data not shown).

Our data may have been subject to misclassification errors in both exposure assessment and case ascertainment. Some vaccinations, particularly the neonatal HBV dose, may not have been captured completely. Mullooly et al[33] evaluated reliability of automated vaccination data in the VSD and estimated that 18% and 2% of HBV may have been missed at HMOs A and B, respectively. For other TCVs, the proportions missed were estimated to be 2% for both diphtheria-tetanus-whole cell pertussis and *Haemophilus influenzae* type b at HMO B and 10% and 9% for the same respective vaccines at HMO A. No specific evaluations of the accuracy of the automated records have been conducted at HMO C, but the accuracy is believed to be high, as the computerized records represent the sole medical record.

For case ascertainment, we used ICD-9 codes at 2 HMOs and Costar codes at the third. The low confirmation rates for ADD illustrate the potentially low positive predictive value of these codes, which could have limited our ability to find an association with this outcome. For other disorders, such as autism, the confirmation rate of the computerized codes was reasonably good. In a subanalysis (not shown), we found consistent results based on computerized codes compared with analyses based on a smaller sample of subjects with autism, ADD, and speech and language disorders whose medical records were reviewed and diagnoses confirmed, suggesting that the reliance on automated data did not introduce appreciable bias.

We were not able to control completely for potentially confounding factors. Clinic identity was unavailable from HMOs A and C and therefore could not be controlled for in the analysis. The variable that denoted which clinic a child attended acted as an appreciable confounder in the analyses at HMO B, and its absence from the other analyses represents a legitimate concern. In terms of the ability for this study to address the effect of other, potentially confounding environmental influences, the HMO databases did not contain information on potential predisposing factors for neurodevelopmental disorders, such as maternal smoking, lead exposure, or fish consumption. However, it is not obvious how these factors would be related to the child's vaccination status and thus confound the results.

LBW is a particularly important potentially confounding factor because LBW infants (especially those severely premature) are less likely to be vaccinated on time,[34] and they are also at increased risk for neurodevelopmental disorders.[35] We dealt with this potential bias by excluding LBW infants from the main analysis. Because LBW infants may be especially susceptible to thimerosal exposure as a result of their higher exposure doses relative to weight and their less developed nervous systems, we performed a subanalysis restricted to infants with moderately low birth weights (1500–2499 g). We were able to evaluate ADD and speech or language disorders and did not find significant increased risks associated with increasing thimerosal exposure.

CONCLUSIONS

In our analyses of computerized HMO data, we found no consistent significant associations between TCVs and neurodevelopmental outcomes. In the first phase of our study, we found an association between exposure to Hg from TCVs and some of the neurodevelopmental outcomes screened. In the second phase, these associations were not replicated for the most common disorders in an independent population. Although the lack of consistency between the 2 phases argues against a thimerosal effect, we believe that additional investigation is required because of the widespread exposure from vaccinating virtually the entire birth cohort of the United States and the

importance of speech and language disorders among children and adolescents. For elucidating further whether a causal association exists between thimerosal exposure and neurodevelopmental conditions, additional studies with different designs will be needed. A study with neuropsychological testing of children with different thimerosal exposures would address one of the main limitations of our present study: the reliance on administrative medical records for outcome assessment. Although such a study might also avoid ascertainment bias that may have affected the results of this study, it might still be susceptible to confounding if factors that influence parents' decision to have their children vaccinated timely are also related to their children's neurodevelopment. Although this bias could conceivably be eliminated by conducting a randomized controlled trial, such a trial would not be ethically feasible given current recommendations that thimerosal not be included in routine infant vaccines. The best alternative is to evaluate the development of children who were enrolled in previous randomized vaccine trials in which the vaccines contained similar antigens but differed by thimerosal content.

APPENDIX 1. Perinatal Exclusion Codes Used in the Thimerosal Screening Analyses

740.*	Anencephalus, craniorachischisis, iniencephaly
741.*	Spina bifida
742.*	Encephalocele, microcephlus, other brain and spinal cord anomalies
745.*	Cardiac defects including ventricular septal defect
746.*	Other congenital heart defects
747.*	Anomalies of aorta, other arteries, veins
748.*	Various abnormalities of nose, lung, respiratory abnormalities
749.*	Cleft palate and cleft lip
750.*	Tongue and mouth abnormalities
751.*	Abnormalities of intestine, pancreas, other digestive
753.0	Renal agenesis
756.6	Anomalies of diaphragm
756.7	Abdominal wall abnormalities
758.*	Chromosomal abnormalities
759.7	Multiple congenital anomalies not elsewhere coded
759.9	Congenital anomaly not otherwise specified
760.*	Maternal condition affecting fetus including maternal injury, hypertension, drugs
761.*	Maternal complication affecting newborn including premature rupture of membrane
764.*	Slow fetal growth, malnutrition, light for gestational age
765.*	Disorders related to short gestation and unspecified low birth weight
767.*	Birth trauma including scalp injury
768.*	Intrauterine asphyxia, fetal distress during labor
769.*	Respiratory distress syndrome
770.*	Newborn respiratory condition
772.1	Intraventricular hemorrhage
772.2	Subarachnoid hemorrhage
773.*	Newborn hemolytic disease
775.*	Newborn endocrinological disease
776.2	Disseminated intravascular coagulation
779.*	Other perinatal condition including convulsion (.0), feeding problems (.3)

Note: 760.* was not used as an exclusion at HMO C.

APPENDIX 2. HMO B: Mean Number of Outpatient Visits for URI (ICD 460–466) and Well-Child Visits (V20*, V70.0, V70.3, V70.5, V70.9) in Second Year of Life by Year of Birth and Estimated Amount of Hg Received In First 7 Months of Life (0–75 vs >75 μg)

Year	μg	*N*	URI				Well-Child Visit			
			Mean	SD	*t* Test	*P* Value	Mean	SD	*t* Test	*P* Value
1994	0–75	2757	1.76	1.93	−4.78	<.0001	1.99	1.26	−15.6	<.0001
	≥75	11 072	1.96	2.00			2.42	1.36		
1995	0–75	2660	1.59	1.77	−3.56	.0004	1.90	1.18	−14.2	<.0001
	≥75	11 405	1.73	1.84			2.26	1.20		
1996	0–75	2563	1.60	1.76	−5.48	<.0001	1.79	1.08	−15.6	<.0001
	≥75	12 059	1.82	1.94			2.16	1.14		
1997	0–75	1222	1.45	1.71	−5.72	<.0001	1.76	1.03	−11.6	<.0001
	≥75	13 486	1.75	1.82			2.12	1.11		
1998	0–75	947	1.60	1.81	0.06	.9540	1.83	1.06	−7.9	<.0001
	≥75	13 643	1.60	1.75			2.12	1.13		

SD indicates standard deviation.
* Children followed continuously >2 years since birth.
Note: Restricting to children with ≤9 visits did not change the results.

APPENDIX 3. RR Associated With Cumulative Hg Exposure by 3 Months of Age for Selected Outcomes for Children Receiving Separate DTP/DTaP and Hib Vaccines Compared with Children Receiving Combined Vaccines, HMO B

Outcome	*N*	No. of Cases	RR	95% CI
Speech delay	29 990	547	1.04	0.85–1.27
Language delay	29 845	167	1.29	0.91–1.82
ADD	13 985	315	0.70*	0.52–0.95

* $P < .05$.

Note: HMO B started using a combined DTP and Hib vaccine in early 1993 and then replaced it with separate DTaP and Hib vaccinations in 1997. The analysis compared the risk among children who received a DTP (or DTaP) and Hib before 3 months of age in a single combination vaccine with those who received the vaccines separately. The analyses were stratified on sex, year of birth, and clinic but not on month of birth (as a result of the rapid change-over of use of vaccine, there was a lack of overlap by month of birth). The RR is expressed as the higher thimerosal group (separate vaccines) compared with the lower group (combined vaccine).

APPENDIX 4. Effects of Applying Exclusion Criteria

	HMO A 7 Months Speech/ Language Delay			HMO B 7 Months Speech Delay		
	Cases	RR	95% CI	Cases	RR	95% CI
All	1115	0.99	0.96–1.03	3477	1.00	0.98–1.02
I	1056	0.99	0.96–1.03	2866	1.01	0.98–1.03
II	969	1.00	0.96–1.03	2654	1.01	0.99–1.04
III	742	0.99	0.95–1.03	1971	1.01	0.99–1.04
IV	730	0.97	0.93–1.02	1941	1.02	0.99–1.05

I Children not continuously enrolled during the first year of life excluded.
II LBW children excluded.
III Children with any condition in Appendix 1 excluded.
IV Children who received <2 polio vaccinations in the first year of life excluded.

ACKNOWLEDGMENTS

The Vaccine Safety Datalink Team at the time of the study included Frank DeStefano MD, MPH, Robert T. Chen, MD, David Shay, MD, MPH, Philip H. Rhodes, PhD, Thomas Verstraeten, MD, Julianne Gee, MPH, and Catherine Okoro (National Immunization Program, Centers for Disease Control and Prevention, Atlanta, GA); Robert S. Thompson, MD, Lisa A. Jackson, MD, MPH, Robert L. Davis, MD, MPH, William E. Barlow, PhD, Kari Bohlke, ScD, Patti Benson, MPH, Barbara Carste, MPH, Jo Ann Habakangas, BA, Christi Hanson, BA, Minqi Jiang, MS, Paula Lee Poy, BA, Darren Malais, BS, Viviana Rebolledo, BS, Wendy Rogers, BA, David Rubanowice, BS, Dennis Sheeran, MS, Onchee Yu, MS, and Ann Zavitkovsky, MPH, MPA (Group Health Cooperative, Seattle, WA); John P. Mullooly, PhD, Julie E. Maher, PhD, Sheila Weinman, PhD, Lois Drew, BA, Jill Mesa, Kim Olson, Heather Houston, RN, Colleen Chun, MD, Steven Gancher, MD, John A. Pearson, MD, Jerry Slepak, MD, Alan Bauck, BS, Teresa Kimes, MS, Joseph Murphy, BA, Nadia Redmond, MSPH, Karen Riedlinger, BS, Roberleigh Schuler, MS, Carol Sullivan, and Gayle Thomas-Monk (Kaiser Permanente Northwest Region, Portland, OR); Steve B. Black, MD, Henry R. Shinefield, MD, Paula Ray, MPH, Edwin Lewis, MPH, Bruce H. Fireman, MA, Joan Schwalbe, Ajit De Silva, and Patti Hallam (Kaiser Permanente of Northern California, Oakland, CA); Joel I. Ward, MD, Connie M. Vadheim, PhD, Hang Lee, PhD, Ken Zangwill, MD, Eileen Eriksen, MPH, Tracy Zhang, MS, Lennifer Lee, MS, Jennie Jing, MA, Nancy Goff, and Jeffrey Perlman, MD (Center for Vaccine Research Harbor-UCLA Medical Center, Torrance, CA); S. Michael Marcy, MD, and Marlene Lugg, DrPH (Southern California Kaiser Permanente, Los Angeles, CA); M. Miles Braun, MD, MPH, Robert P. Wise, MD, MPH, and Robert Ball, MD, MPH (Center for Biologics Evaluation and Research, Food and Drug Administration, Rockville, MD); and Vito Caserta, MD, MPH, and Geoffrey Evans, MD (Division of Vaccine Injury Compensation, Health Resources and Services Administration, Rockville, MD). We also acknowledge Inna Dashevsky, MS, Stevan deProsse, Ursula Lebron, Richard Platt, MD, MS, and Emily Cain, MPH (Harvard Pilgrim Health Care, Boston, MA).

REFERENCES

1. Ball LK, Ball R, Pratt RD. An assessment of thimerosal use in childhood vaccines. *Pediatrics*. 2001;107:1147–1154
2. Agency for Toxic Substances and Disease Registry. *Toxicological Profile for Mercury* (Update). Atlanta, GA: Agency for Toxic Substances and Disease Registry; 1999
3. Bakir F, Damluji SF, Amin-Zaki L, et al. Methylmercury poisoning in Iraq. *Science*. 1973;181:230–241
4. Igata A. Epidemiological and clinical features of Minamata disease. *Environ Res*. 1993;63:157–169
5. Davidson PW, Myers GJ, Cox C, et al. Longitudinal neurodevelopmental study of Seychellois children following in utero exposure to methylmercury from maternal fish ingestion: outcomes at 19 and 29 months. *Neurotoxicology*. 1995;16:677–688
6. Mahaffey KR, Rice G. *An assessment of Exposure to Mercury in the United States: Mercury Report to Congress*. Washington, DC: US Environmental Protection Agency; 1997
7. Grandjean P, Weihe P, White RF, et al. Cognitive deficit in 7-year-old children with prenatal exposure to methylmercury. *Neurotoxicol Teratol*. 1997;19:417–428
8. WHO. *Environmental Health Criteria: Mercury*. Geneva, Switzerland: World Health Organization; 1976.
9. US Environmental Protection Agency. *Mercury Study Report to Congress. Volume V: Health Effects of Mercury and Mercury Compounds*. Washington, DC: Office of Air Quality Planning and Standards and Office of Research and Development; 1997
10. 21 USC, 397 Section 413; 2001
11. Verstraeten T, Gu D, Chen R. Exposure to Ethylmercury From Thimerosal Containing Vaccines in Infants. Presented at the 127th Annual Meeting of the American Public Health Association; November 9, 1999; Chicago, IL
12. Thimerosal in vaccines: a joint statement of the American Academy of Pediatrics and the Public Health Service. *Neonatal Netw*. 1999;18:65–72
13. Centers for Disease Control and Prevention. Recommendations regarding the use of vaccines that contain thimerosal as a preservative. *JAMA*. 1999;282:2114–2115
14. Centers for Disease Control and Prevention. Availability of hepatitis B

vaccine that does not contain thimerosal as a preservative. *JAMA*. 1999;282:1219–1220
15. Jessen P, Verdiere B, Moriet G, Boule D, Rhaoui A, Fraisse F. [An original mechanism of acute mercury poisoning: ritual injection]. *Presse Med*. 1991;20:1625–1626
16. Lowell JA, Burgess S, Shenoy S, Curci JA, Peters M, Howard TK. Mercury poisoning associated with hepatitis-B immunoglobulin. *Lancet*. 1996;347:480
17. Netscher DT, Friedland JA, Guzewicz RM. Mercury poisoning from intravenous injection: treatment by granuloma resection. *Ann Plast Surg*. 1991;26:592–596
18. Zosin C, Manescu N, Gluhovschi G, Nicolcioiu M, Golea O, Bignion H. [Acute mercury poisoning due to intravenous injection of metallic mercury]. *MMW Munch Med Wochenschr*. 1977;119:1537–1538
19. Chen RT, Glasser JW, Rhodes PR, et al. Vaccine Safety Datalink project: a new tool for improving vaccine safety monitoring in the United States. The Vaccine Safety Datalink Team. *Pediatrics*. 1997;99:765–773
20. Chen RT, DeStefano F, Davis RL, et al. The Vaccine Safety Datalink: immunization research in health maintenance organizations in the USA. *Bull World Health Organ*. 2000;78:186–194
21. Wassilak SG, Glasser JW, Chen RT, Hadler SC. Utility of large-linked databases in vaccine safety, particularly in distinguishing independent and synergistic effects. The Vaccine Safety Datalink Investigators. *Ann N Y Acad Sci*. 1995;754:377–382
22. Crump KS, Van Landingham C, Shamlaye C, et al. Benchmark concentrations for methylmercury obtained from the Seychelles Child Development Study. *Environ Health Perspect*. 2000;108:257–263
23. Grandjean P, White RF, Nielsen A, Cleary D, De Oliveira Santos EC. Methylmercury neurotoxicity in Amazonian children downstream from gold mining. *Environ Health Perspect*. 1999;107:587–591
24. Steuerwald U, Weihe P, Jorgensen PJ, et al. Maternal seafood diet, methylmercury exposure, and neonatal neurologic function. *J Pediatr*. 2000;136:599–605
25. Davidson PW, Myers GJ, Cox C, et al. Effects of prenatal and postnatal methylmercury exposure from fish consumption on neurodevelopment: outcomes at 66 months of age in the Seychelles Child Development Study. *JAMA*. 1998;280:701–707
26. Grandjean P, Budtz-Jorgensen E, White RF, et al. Methylmercury exposure biomarkers as indicators of neurotoxicity in children aged 7 years. *Am J Epidemiol*. 1999;150:301–305
27. Marsh DO, Myers GJ, Clarkson TW, Amin-Zaki L, Tikriti S, Majeed MA. Fetal methylmercury poisoning: clinical and toxicological data on 29 cases. *Ann Neurol*. 1980;7:348–353
28. Grandjean P, Weihe P, White RF, Debes F. Cognitive performance of children prenatally exposed to "safe" levels of methylmercury. *Environ Res*. 1998;77:165–172
29. Stajich GV, Lopez GP, Harry SW, Sexson WR. Iatrogenic exposure to mercury after hepatitis B vaccination in preterm infants. *J Pediatr*. 2000; 136:679–681
30. Pichichero ME, Cernichiari E, Lopreiato J, Treanor J. Mercury concentrations and metabolism in infants receiving vaccines containing thimerosal: a descriptive study. *Lancet*. 2002;360:1737–1741
31. Stratton K, Gable A, McCormick MC, eds. *Immunization Safety Review: Thimerosal-Containing Vaccines and Neurodevelopmental Disorders. Institute of Medicine. Immunization Safety Review Committee*. Washington, DC: National Academy Press; 2001
32. Howson CP, Howe CJ, Fineberg HV, eds. *Adverse Effects of Pertussis and Rubella Vaccines. Institute of Medicine*. Washington, DC: National Academy Press; 1991
33. Mullooly J, Drew L, DeStefano F, et al. Quality of HMO vaccination databases used to monitor childhood vaccine safety. *Am J Epidemiol*. 1999;149:186–194
34. Davis RL, Rubanowice D, Shinefield HR, et al. Immunization levels among premature and low-birth-weight infants and risk factors for delayed up-to-date immunization status. *JAMA*. 1999;282:547–53
35. Anderson P, Doyle LW. Neurobehavioral outcomes of school-age children born extremely low birth weight or very preterm in the 1990s. *JAMA*. 2003;289:3264–3272

MARGARET MEAD'S EARLY EDUCATION

"Kindergarten passed muster among the pedagogically enlightened women of the family. Margaret attended for two years. But from then until fourth grade, she had no official schooling. When she finally did show up in class it was only for half days, and she went armed with instructions from her parents that she was to be permitted to leave whenever she liked. When nine-year old Margaret decided to begin a diary, her first entry showed her to be very much the product of her eclectic yet ever so self-conscious rearing: 'I'm not sure that I won't miss days some times, for I am not very regalar.' At adolescence, she was swept up in a joint project with her mother . . . the two of them went off to study Italian immigrant children, aiming to find out how language affected IQ scores."

Hulbert A. *Raising America*. New York: Knopf; 2003

Submitted by Student