

Reye's Syndrome and Aspirin

Evidence for a Dose-Response Effect

Paul F. Pinsky, MPH; Eugene S. Hurwitz, MD;

Lawrence B. Schonberger, MD, MPH; Walter J. Gunn, PhD

Data collected from the Public Health Service Main Study of Reye's Syndrome and Medications were analyzed to assess the relationship between the development of Reye's syndrome and the dose of aspirin received during the antecedent respiratory or chickenpox illness. Among those exposed to aspirin, case-patients were found to have received greater average daily and maximum daily doses of aspirin and greater doses of aspirin on the first four days of the antecedent illness (median, 25.1 mg/kg; 33.0 mg/kg; and 65.4 mg/kg; respectively) than did controls (median, 14.5 mg/kg; 19.0 mg/kg; and 27.0 mg/kg; respectively). The excess risk associated with increasing aspirin doses was due primarily to intermediate levels of dose (eg, 15 to 27 mg/kg per day) rather than higher levels (>27 mg/kg per day). The dose difference between exposed case-patients and controls was greatest on days 3 and 4 of the antecedent illness.

(*JAMA* 1988;260:657-661)

SINCE 1980, six studies,^{1,5} including the recently reported Public Health Service (PHS) Main Study of Reye's Syndrome and Medications, have been reported that demonstrated a strong association between Reye's syndrome and taking salicylates (specifically aspirin) during the antecedent respiratory or chickenpox illness that typically precedes the disease. In addition to the tendency for case-patients to receive aspirin more frequently, a trend for such patients to receive higher doses of aspirin than controls was reported in three of the four previous studies that examined dosage.¹⁻³ In this report we describe analyses and results of the recently reported PHS Main Study of Reye's Syndrome and Medications designed to explore further the nature of the rela-

tionship between dose of aspirin and Reye's syndrome. We compare various characteristics of the aspirin exposure among cases and controls, including the relationship between risk of Reye's syndrome and timing, duration, and dose of aspirin administered during the antecedent illness, and quantify increases in risk associated with different aspirin dose levels. In addition to providing supportive epidemiologic evidence for the causal association between Reye's syndrome and aspirin, these analyses may provide further clues to the pathophysiology of this illness.

METHODS

The methods used in the PHS Main Study of Reye's Syndrome and Medications have been described elsewhere.⁵ The study involved a comparison of medications administered to patients with Reye's syndrome (identified from 70 pediatric tertiary-care centers) during antecedent respiratory, chicken-

pox, or gastrointestinal illnesses with medications administered to control children with similar illnesses. Four different control groups were compared with Reye's syndrome cases—emergency department, inpatient, school, and community. Due to the relatively small numbers of controls in each group who used aspirin (8, 4, 16, and 12, respectively), the control groups were aggregated in our analysis.

Three types of care providers (main care providers, other care providers, and the subjects themselves whenever possible) were questioned about the medications administered during the subject's antecedent illness. A given administration of a specific medication to a study child, reported by one of these care providers, was defined as a *medication event*; if the medication in question contained aspirin, then we labeled this an *aspirin medication event*. These care providers were asked to list all medication events that they were aware of and for each one to give the date of the event, the exact (brand) name of the medication involved, the name of the individual who administered the medication, and the total number of standard dosage units administered. A *standard dosage unit* was defined as a tablet, tablespoon, or other unit of measurement (eg, drop) appropriate to the physical form of the medication. Only those medication events that were reported by the care provider who actually administered the medication were used in our analysis.

For each medication mentioned, a pharmacist calculated the number of

From the Division of Viral Diseases, Center for Infectious Diseases, Centers for Disease Control, Atlanta.
Reprint requests to Centers for Disease Control, Bldg 6, Room 119, Atlanta, GA 30333 (Mr Pinsky).

Table 1.—Characteristics of Case-Patients and Controls Who Received Aspirin

Characteristic	Case-Patients, No. (%)	Controls, No. (%)
Age, y		
0-6	3 (12)	2 (6)
7-10	6 (24)	10 (31)
11-14	12 (48)	14 (44)
15-18	4 (16)	6 (9)
Total	25 (100)	32 (100)
Nonblack race	24 (96)	31 (97)
Respiratory illness	23 (92)	31 (97)

Table 2.—Characteristics of Medication Events of Case-Patients and Controls Who Received Aspirin

Aspirin Administration	Case-Patients (n=25)	Controls (n=32)	P*
No. of aspirin medication events (days 1-4)†			
Median (range)	5.0 (1-14)	3.0 (1-16)	<.05
Mean	6.0	4.6	
Average daily No. of aspirin medication events			
Median (range)	2.0 (1-4.0)	1.2 (0.5-4.0)	<.01
Mean	2.0	1.5	
No. of days exposed to aspirin (days 1-4)			
Median (range)	3 (0-4)	2 (0-4)	.01
Mean	2.7	1.9	

*Wilcoxon rank-sum test.

†Includes only those 24 cases and 26 controls exposed during days 1 to 4.

Table 3.—Characteristics of Aspirin Dose Among Case-Patients and Controls Who Received Aspirin

Dose Measure	Dosage, mg/kg		P*
	Case-Patients (n=23)	Controls (n=31)	
Average daily			
Median (range)	25.1 (6.1-71.2)	14.5 (4.2-55.5)	<.01
Geometric mean	25.8	15.4	
Maximum daily			
Median (range)	33.0 (6.1-94.9)	19.0 (4.2-56.3)	.001
Geometric mean	31.8	17.3	
4 Day†			
Median (range)	65.4 (6.1-284.8)	27.0 (4.2-204.1)	<.01
Geometric mean	61.6	29.6	

*t test.

†Includes only those 22 cases and 26 controls exposed during days 1 to 4.

milligrams of each of the active ingredients per standard dosage unit. All dosages referred to in the analysis are expressed as milligrams per kilogram of body weight.

In all our analyses for cases, we considered only those exposures that occurred during the antecedent illness before the clinically defined onset of Reye's syndrome. For controls, unless otherwise stated, we examined all exposures reported during their comparable illness.

To examine possible differences in the pattern of aspirin doses received by cases vs controls we looked at the following three measures of dose: average daily dose (total aspirin dose divided by number of days aspirin was administered), maximum daily dose (maximum aspirin dose administered in a single

day), and four-day dose (total aspirin dose on the first four days of the antecedent or comparable illness). This latter measure was used as a surrogate for total dose to ensure that cases and controls had comparable exposure intervals. On average, cases received 85% of their total aspirin dose on days 1 to 4 and only one exposed case did not receive aspirin by day 4. When examining four-day dose we limited our analyses to those exposed during days 1 to 4.

In addition, we examined certain indirect indexes of aspirin dose: these included average daily number of aspirin medication events, number of aspirin medication events on the first four days, and the number of days, through day 4, exposed to aspirin. The Wilcoxon rank-sum test was used to assess statistical significance regarding the indirect

dose indexes while Student's *t* test (based on log dose) was used for the direct dose measures. Weights were unavailable for two cases and one control; therefore, their milligram per kilogram doses could not be calculated. These subjects were used, however, in analyzing the indirect dose measures.

Multivariate analysis using conditional logistic regression models was employed to obtain estimates of risk associated with different levels of dose and to control for the effect of matching variables. The original protocol for the PHS Main Study of Reye's Syndrome and Medications specified a study design in which each case was individually matched to several controls. For the purposes of analyzing dose in this study, it was more efficient to use group matching; this enabled us to utilize data on all exposed individuals while retaining the essentials of the matched design.⁶ Group matching on age was done according to the categories shown in Table 1. The other two original matching variables (race and type of antecedent illness) had limited disparity in both cases and controls (>90% in each group were nonblack and had respiratory illness); thus, these were not included in the group matching criteria.

Each regression model treated the development of Reye's syndrome as the dependent variable and included a dichotomous variable, which indicated aspirin exposure as well as a variable(s) representing one of the direct dose measures as independent variables. All subjects, both unexposed and exposed, were included in these models.

In our first set of logistic models we classified each dose as lower, intermediate, or higher based on the overall distribution of that dose measure in cases and controls. This enabled us to estimate the odds ratios of different levels of exposure relative to other levels of exposure or to no aspirin exposure.

To corroborate the validity of these findings and to gain further insight into the shape of the dose-response curve, we also developed continuous models using both dose and log dose as the independent variables.

As noted in the report of the main study, the severity of the antecedent illness, as measured by the severity index, was in general greater for controls than for cases. Therefore, we chose not to include the severity index covariates in the model. Including these variables would have resulted in an increase in the observed odds ratios.

The statistical significance of all findings was assessed using one-tailed tests unless otherwise noted.

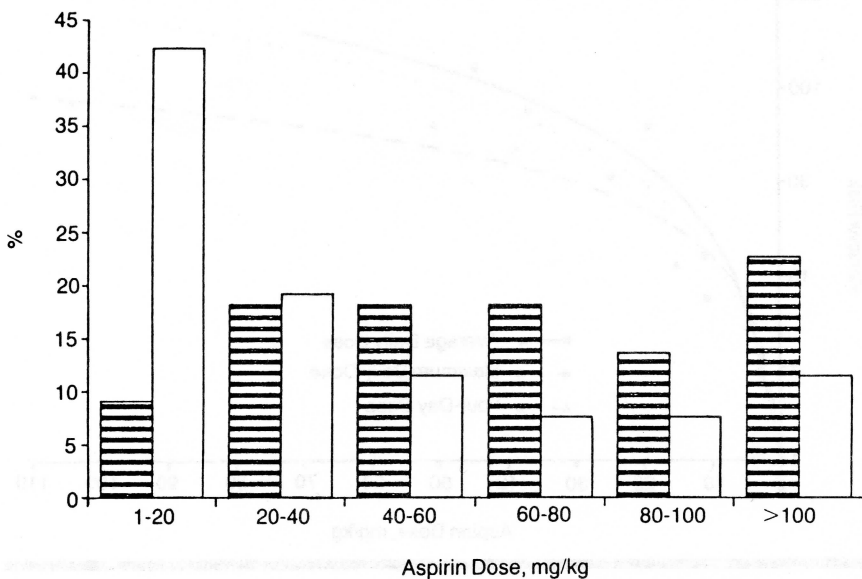


Fig 1.—Distribution of four-day aspirin dose in cases and controls receiving aspirin by day 4. Striped bars indicate case-patients; and hollow bars, controls.

Table 4.—Logistic Model—Estimates of Relative Risk for Different Dose Levels of Aspirin

Dose Measure of Aspirin	Dose Range, mg/kg	Relative Risk*	Incremental Risk†		95% Confidence Interval
			Risk	P	
Average daily					
Lower	4-15	7.3	7.3	.03	1.2-42.2
Intermediate	15-27	61.4	8.5	<.01	2.0-36.1
Higher	27-71	73.6	1.2	NS	0.4-3.6
Maximum daily					
Lower	4-18	12.4	12.4	<.01	2.4-64.2
Intermediate	18-33	33.3	2.7	NS	0.7-10.0
Higher	33-95	126.4	3.8	.03	1.2-12.4
4 Day					
Lower	4-28	11.0	11.0	.01	2.0-60.0
Intermediate	28-69	61.8	5.6	.02	1.4-23.0
Higher	69-285	83.5	1.4	NS	0.4-4.4

*Relative risk of given level of exposure relative to unexposed.

†Relative risk of given level of exposure relative to next lower level of exposure (eg, lower:unexposed, intermediate:lower, and higher:intermediate).

RESULTS

Care providers reported directly administering aspirin containing medications during the subject's antecedent illness to 93% (25/27) of case-patients and to 23% (32/140) of controls. (For eight additional controls, aspirin medication events were reported only by individuals who did not administer the medication.)

As seen in Table 1, exposed cases and controls were generally similar with respect to age (mean age, 11.1 years and 11.5 years, respectively).

The total number of aspirin medication events on the first four days, as well as the average daily number of aspirin

medication events, was found to be significantly elevated in cases compared with controls (Table 2). When the total and average numbers of all medication events were examined, however, no significant differences between cases and controls were found. Cases were also found to have been exposed on significantly more of the first four days of the antecedent illness than were controls.

As seen in Table 3, the (geometric) mean of each dose measure (average, maximum, and four day) was significantly greater in cases than in controls. Fig 1 displays the distribution of four-day dose in cases and controls exposed during this period. Seventy-three per-

cent of the case-patients were exposed to at least 40 mg/kg of aspirin during the first four days of the antecedent illness compared with only 39% of the controls. Similarly, 87% of the case-patients were exposed to at least 15 mg/kg per day of aspirin compared with only 45% of the controls.

Logistic Regression Models

Table 4 gives estimates of the relative risk of each dose level of aspirin compared with no exposure and compared with the next lower level of exposure; we term this latter measure the *incremental risk*.

As seen from Table 4, lower dose exposure to aspirin vs no aspirin exposure was associated with a significantly elevated relative risk ranging from 7.3 for average daily dose to 12.4 for maximum dose. Exposure to intermediate and higher doses of aspirin vs no aspirin exposure, however, was associated with a markedly greater relative risk; thus the relative risk of an intermediate average daily dose of aspirin was 61.4 and the relative risk of a higher average daily dose was 73.6.

Individuals exposed to intermediate four-day aspirin doses were 5.6 times more likely (ie, incremental risk of 5.6) to develop Reye's syndrome than those exposed to lower four-day doses and 61.8 times more likely (ie, relative risk of 61.8) to develop Reye's syndrome than those not exposed to aspirin. Intermediate average daily doses of aspirin also carried significantly elevated incremental risk (8.5). The incremental risk for intermediate (compared with lower) maximum daily doses was relatively low (2.7) and not significantly different from 1; however the incremental risk of higher compared with intermediate maximum daily doses was significantly elevated (3.8). The estimated incremental risk for higher compared with intermediate doses was minimal for average daily dose and four-day dose; neither of these incremental risks were significantly different from 1. For each dose measure, however, the relative risk of higher compared with lower dose exposure did significantly exceed 1, ranging from 7.6 for four-day dose to 10.2 for maximum daily dose (not shown in Table 4).

Each of the continuous models demonstrated a significant effect of aspirin dosage on the risk of Reye's syndrome among those exposed to aspirin. Analysis of the goodness of fit of the continuous models demonstrated that the log model outperformed the linear model for all three dose measures.⁷ Comparison of the log-dose model with the categorical model provides further support

for the validity of the log-dose model. Estimates of the log relative risk of low, moderate, and high aspirin doses generated from the categorical models, when plotted at the median observed dose within each category, closely approximated the estimates generated using the log-dose models (Fig 2).

Dose by Day

We attempted to characterize further the observed dose increase in cases compared with controls by examining dose on specific days of the antecedent illness. For each of the first four days of the antecedent illness, a significantly higher proportion of case-patients were exposed to aspirin (67%, 78%, 78%, and 73%, respectively) than were controls (14%, 14%, 9%, and 9%, respectively). As seen in Fig 3, among those who received aspirin on a given day, aspirin doses on days 1 and 2 were similar in cases and controls; however, case-patients received significantly higher doses of aspirin than did controls on days 3 and 4.

COMMENT

These findings suggest that in addition to the strong association between exposure to aspirin and development of Reye's syndrome observed in this study and several prior studies, larger doses of aspirin (though generally, as in previous studies, well below the recommended upper limit for antipyretic therapy of 80 mg/kg per day⁸) administered during the antecedent illness are related to an increased risk of disease. Although considerable overlap in doses was observed, case-patients tended to receive higher doses of aspirin during the first four days, and higher doses of aspirin on each day exposed, than did controls. Such an epidemiologic observation lends strong additional support for a causal role of aspirin in the development of Reye's syndrome.

The results of this study suggest that even in low doses aspirin is a significant risk factor for developing Reye's syndrome. Exposure to as little as 15 mg/kg per day of aspirin (eg, about two 325-mg tablets for a 40-kg child) was associated with a sevenfold increase in the risk of developing Reye's syndrome. It should be noted, however, that the doses reported herein may underestimate the true dose since only those medication events reported by the individual who actually administered the medication were utilized in this analysis (these accounted for 71% and 70% of the medication events reported for case-patients and controls, respectively).

Too few subjects (four cases and 14

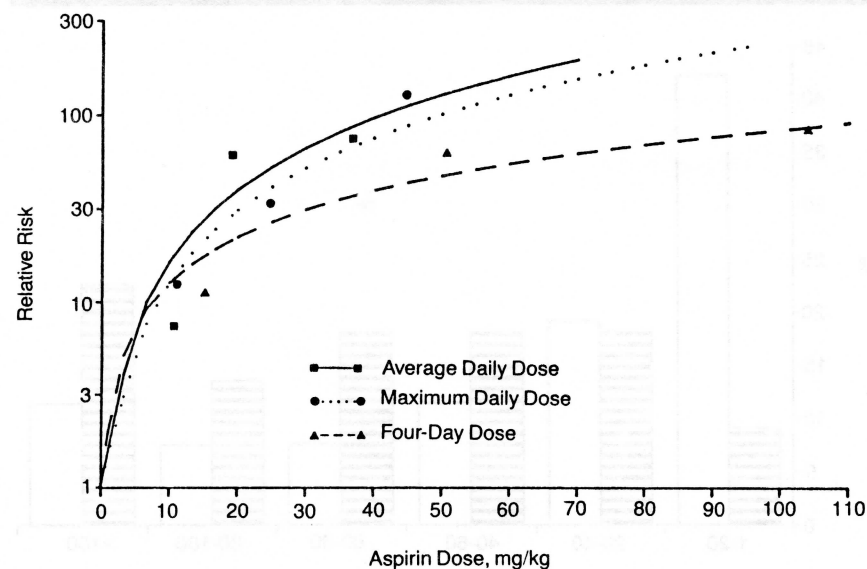


Fig 2.—Curves of log-relative risk vs aspirin dose. Symbols represent relative risk estimates for lower, intermediate, and higher dose levels plotted at median observed dose within given level. Lines represent relative risk curves derived from continuous logistic models using log dose.

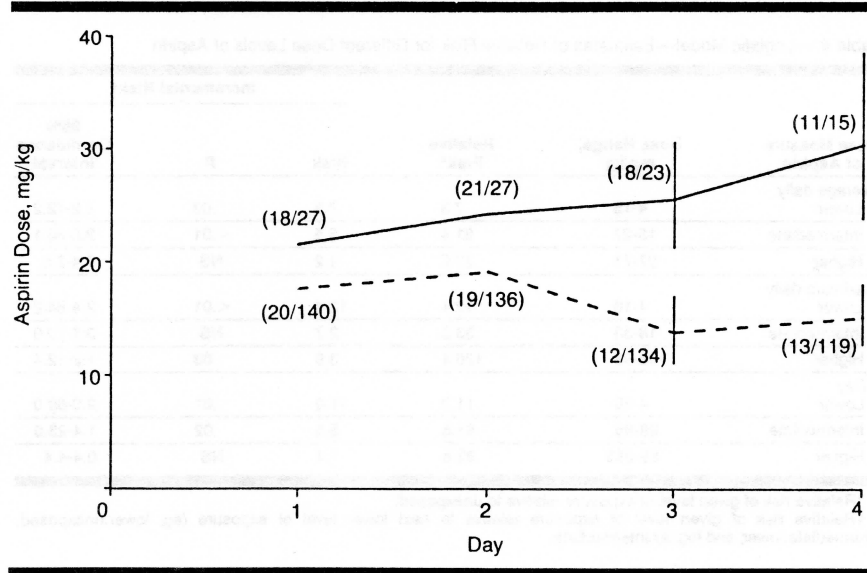


Fig 3.—Aspirin dose by day: case-patients vs controls. Numerators of fractions in parentheses give number exposed on given day. Denominators give number of cases who have not yet developed Reye's syndrome or controls who are still ill. Error bars (± 1 SEM) denote significant ($P < .05$) differences in geometric mean dose between cases and controls. Dashed line indicates controls; and solid line, case-patients.

controls) were exposed to nonaspirin salicylates, which included subsalicylate bismuth, salicylate magnesium, and salicylamide, to assess these compounds independently in terms of a possible dose-response relationship with Reye's syndrome. The median total dose of salicylate from nonaspirin sources was 5.9 mg/kg in cases compared with 4.5 mg/kg in controls.

To address concerns related to a possible tendency of case respondents to recall better the quantity of medication

doses that they administered, indirect indexes of aspirin dose (eg, number of aspirin medication events and number of days exposed to aspirin), which are not as dependent on the numerical accuracy of respondents' recall as are the direct dose measures, were assessed. These indexes were found to be significantly higher in cases than in controls.

The milligram per kilogram doses of aspirin reported herein for Reye's syndrome cases are similar to those reported in earlier studies.²⁴ Only in the Michi-

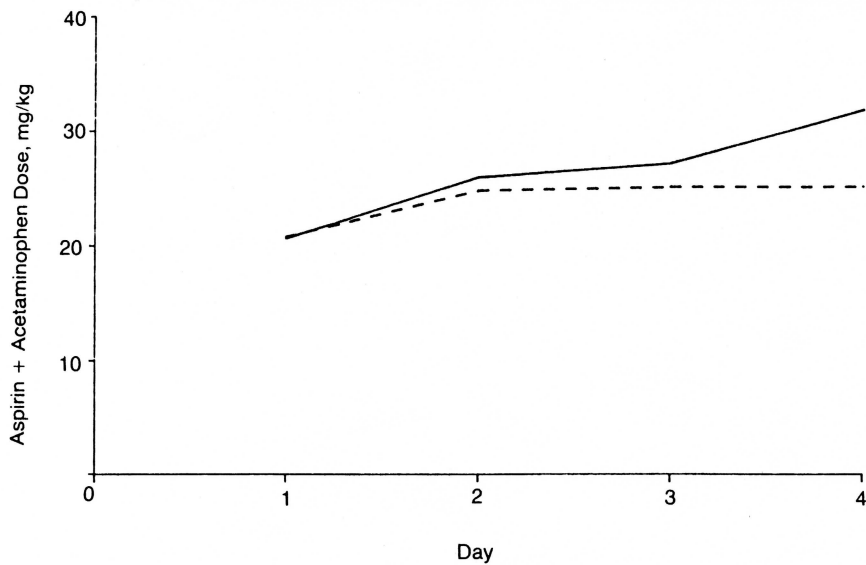


Fig 4.—Combined aspirin and acetaminophen dose by day: case-patients vs controls. Dashed line indicates controls; and solid line, case-patients.

analysis was that among exposed subjects, case-patients received significantly higher doses of aspirin than did controls on days 3 and 4 of the antecedent illness; no differences in dosage between case-patients and controls were noted on days 1 and 2, however. One possible explanation for this finding would be a differential daily pattern of illness in cases compared with controls (ie, increasing severity by day in case-patients and/or decreasing severity by day in controls). This hypothesis, however, is not supported by either the parents' subjective evaluation of illness severity or by data on nonaspirin medications. For instance, Fig 4 shows the combined dose of aspirin plus acetaminophen for days 1 through 4; no significant differences in dose were observed.

Particular attention concerning the pathophysiological process leading to Reye's syndrome has focused on metabolic pathways in the mitochondria. It is possible that aspirin, in conjunction with certain viral infections, has a dose-dependent effect on a certain mitochondrial enzyme (or enzymes) leading to this disease, perhaps in genetically predisposed children. Though animal or in vitro models have not been fully successful, the fact that low-dose aspirin administration in the presence of viral infection can inhibit a mitochondrial enzyme and that aspirin can act in a dose-dependent manner in causing mitochondrial injury have been documented in laboratory studies.^{9,10}

Given our current understanding of the Reye's syndrome-aspirin relationship, it must be assumed that no safe dose of aspirin exists and that avoidance of this compound for treating children and teenagers with chickenpox or respiratory illness is the most effective means of reducing the risk of developing this illness.

The results of the logistic analysis described herein are consistent with a dose-response curve that has a steep slope at lower to intermediate aspirin dose levels and a flatter slope at higher dose levels (within the range observed in this study). The relative risk of lower four-day and average daily doses and the incremental risk of intermediate compared with lower four-day and average daily doses were significantly greater than 1, whereas the incremental risk of higher compared with intermediate four-day and average daily doses did not differ significantly from 1. The results of the continuous logistic models also tend to support the idea of a relative flattening of the dose-response curve at higher dose levels.

Another significant finding in this

gan study² were control weights obtained; there the mean total, average, and maximum daily milligram per kilogram doses were markedly higher in cases ($n = 12$) than in controls. Starko et al¹ reported a trend for cases ($n = 7$) to receive higher milligram doses of aspirin than controls in a study in Arizona. In the Ohio study,³ analyses were completed comparing cases and controls stratified for age at five-year intervals; the average and maximum daily doses in milligrams were found to be significantly elevated in cases compared with controls. In the PHS pilot study, milligram per kilogram doses of aspirin were estimated based on age and sex. In this study the average daily aspirin doses in cases (mean, 21.9 mg/kg) and controls (mean, 18.3 mg/kg) were similar.

References

1. Starko KM, Ray CG, Dominguez LB, et al: Reye's syndrome and salicylate use. *Pediatrics* 1980;66:859-864.
2. Waldman RJ, Hall WN, McGee H, et al: Aspirin as a risk factor in Reye's syndrome. *JAMA* 1982;247:3089-3094.
3. Halpin TJ, Holtzauer FJ, Campbell RJ, et al: Reye's syndrome and medication use. *JAMA* 1982;248:687-691.
4. Hurwitz ES, Barrett MJ, Bregman D, et al: Public Health Service study of Reye's syndrome and medications: Report of the pilot phase. *N Engl J Med* 1985;313:849-857.
5. Hurwitz ES, Barrett MJ, Bregman D, et al: Public Health Service study of Reye's syndrome and medications: Report of the main study. *JAMA* 1987;257:1905-1911.
6. Brookmeyer R, Liang K, Linet M: Matched case-control designs and overmatched analyses. *Am J Epidemiol* 1986;124:693-700.
7. Lemeshow S, Hosmer DW: A review of goodness of fit statistics for use in the development of logistic regression models. *Am J Epidemiol* 1982;115:92-106.
8. Vaughan VL, McKay RJ, Behrman RE: *Nelson Textbook of Pediatrics*, ed 11. Philadelphia, WB Saunders Co, 1979.
9. Trauner DA, Horvath E, Davis LE: Inhibition of fatty acid beta oxidation by influenza B virus and salicylic acid in mice: Implications for Reye's syndrome. *Neurology* 1988;38:239-241.
10. You K: Salicylate and mitochondrial injury in Reye's syndrome. *Science* 1983;221:163-165.