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ORIGINAL REPORT

Effects of gestational age at enrollment in pregnancy exposure registries†

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ABSTRACT

Purpose This study aims to explore the influence of gestational age at enrollment, and enrollment before or after prenatal screening, on the estimation of drug effects in pregnancy exposure registries.

Methods We assessed the associations between first trimester antiepileptic drug (AED) exposure and risk of spontaneous abortion and major congenital malformations in the North American AED Registry (1996–2013). We performed logistic regression analyses, conditional or unconditional on gestational age at enrollment, to estimate relative risk (RR) for first trimester AED users compared with non-users. We also compared first trimester users of valproic acid and lamotrigine. Analyses were repeated in women who enrolled before prenatal screening.

Results Enrollment occurred earlier among 7029 AED users than among 581 non-users; it was similar among AEDs. Comparing AED users with non-users, RR (95% confidence interval) of spontaneous abortion ($n = 359$) decreased from 5.1 (2.3–14.1) to 2.0 (0.9–5.6) after conditioning on gestational week at enrollment and to 1.9 (0.8–5.4) upon further restriction to before-screening enrollees. RR of congenital malformations ($n = 216$) changed from 3.1 (1.4–8.5) to 3.2 (1.4–9.0) after conditioning on gestational week at enrollment and to 2.0 (0.7–10.1) upon further restriction to before-screening enrollees. When comparing valproic acid users and lamotrigine users, the RR of congenital malformations was not substantially changed by conditioning or restricting.

Conclusions Spontaneous abortion rates were sensitive to gestational age at enrollment. Estimates of congenital malformation risks for AED users relative to non-users were sensitive to before/after-screening enrollment. This difference was not apparent between active drugs, likely due to similar gestational age at enrollment. Copyright © 2015 John Wiley & Sons, Ltd.

KEY WORDS—pregnancy registries; left truncation; anticonvulsants; spontaneous abortions; major congenital malformations; pharmacoepidemiology

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INTRODUCTION

Pregnant women are generally excluded from the clinical trials that generate pre-approval human data.[1] However, once approved, drugs are available to women who are (or will become) pregnant during treatment. Pregnancy exposure registries are the most efficient source of safety information on new drugs that are likely to be used by pregnant women[2] and a major source of information on existing drugs.[2,3]

Pregnancy exposure registries should enroll participants as early as possible in order to prospectively document risk factors and early-pregnancy events such as spontaneous abortions.[4] When a spontaneous abortion occurs before enrollment and the pregnancy is lost to the registry, we are in the presence of left truncation. Early enrollment also ensures that participation is not prompted or hindered by pregnancy complications or by the results of prenatal screening tests.[5,6]

Pregnant women may seek enrollment in a registry at any time during gestation. Pregnancy registry guidelines recommend inclusion only of participants enrolled prior to prenatal screening[3,5,7] but do not offer detailed guidance on handling the varying gestational age at enrollment and the resulting dynamic study size changes in analyses. Researchers have used analytical methods that incorporate varying gestational age at the beginning of follow-up (delayed or staggered entry, or left truncation) to study spontaneous abortion. However, whether enrollment occurs before or after prenatal screening has not generally been considered.[8–10]

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In the present study, we explore the effects of left truncation on the estimates from a pregnancy registry. Concretely, we evaluate the influence of gestational age at enrollment, and enrollment before and after prenatal screening, on the results from pregnancy exposure cohorts taken from the North American Antiepileptic Drug (AED) Registry. We use as examples the associations between antiepileptic drug use and two pregnancy outcomes: spontaneous abortion and major congenital malformations.

METHODS

Data source

The North American AED Pregnancy Registry[6,11–13] was established in 1996 to provide post-marketing information on the association between antiepileptic drug use in pregnancy and major congenital malformations in exposed infants. Funded by drug manufacturers, it is run independently at a university-affiliated hospital.

Pregnant women in the USA and Canada taking antiepileptic drugs enroll by calling a toll-free number in drug inserts, epilepsy-related Websites, and printed material. After giving verbal informed consent, women are administered a computer-assisted telephone interview asking about antiepileptic drug use, indications, and medical history. Women who enroll before prenatal screening for congenital anomalies (nuchal translucency measurements, chorionic villous sampling, amniocentesis, maternal serum analyte measurements, cell free fetal DNA sequencing, and second-trimester ultrasound) are classified as "pure prospective" (herein, before-screening) enrollees; others are classified as "traditional prospective" (herein, after-screening) enrollees. In this study, women who enrolled after gestational week 20 were considered after-screening enrollees even in the few instances where they reported no prenatal screening (1% of the study population), because we assumed that women have likely had some prenatal screening by then in the context of health care in North America.

The second computer-assisted telephone interview takes place in the seventh month of pregnancy and the third one 8–12 weeks after the expected due date. If a woman enrolled in the registry calls to report having had a spontaneous abortion, elective termination, or stillbirth, she is offered to complete the seventh month interview, which includes questions on changes in treatment, health, and lifestyle since the first interview and information related to the ending of the pregnancy, if appropriate. In addition, she would be asked for any relevant study results, such as chromosome analysis or an autopsy. Enrollees are asked to authorize access to their medical records; approximately 65% agree. A clinical teratologist from the North American AED Pregnancy Registry (L. B. H.) reviews those records for further information on offspring health.

In 2003, to allow comparison of antiepileptic drug users to non-users within the registry, enrollment was opened to unexposed pregnant women who were friends or relatives of exposed enrollees. Non-users undergo the same enrollment and interview process as antiepileptic drug users.

Chromosomal and single-gene abnormalities, as well as minor anomalies, birth marks, and complications of prematurity, are excluded from analyses of malformations in the registry.[14] Women who withdrew from the registry or were lost to follow-up were excluded from these analyses. Those whose gestational age at enrollment, before/after-screening status, or pregnancy outcome were unknown were also excluded. This study was approved annually by the Human Studies Committee of the Massachusetts General Hospital and Partners Health Care.

Exposure and outcomes

Women who reported having used one or more antiepileptic drugs within 3 months after the first day of their last menstrual period were considered exposed; the remaining women were considered unexposed. The drugs are listed as a footnote to Table 1. We evaluated all antiepileptic drugs combined and lamotrigine and valproic acid separately. We selected lamotrigine and valproic acid because the former is considered relatively safe in pregnancy, while the latter is considered more hazardous.[15–17]

A spontaneous abortion was defined as spontaneous loss of pregnancy between gestational weeks 5 and 20. Major congenital malformations were defined as structural malformations of medical, surgical, or cosmetic relevance[18] reported at or before the third interview. All live births, stillbirths, and fetuses from electively terminated pregnancies were included. However, because we did not have relevant information on embryos or fetuses from spontaneous abortions, these were excluded from analyses of major congenital malformations.

Statistical analysis

Gestational age at enrollment by exposure status. We plotted the percentage of participants enrolled each gestational month among users of any antiepileptic drug, users of lamotrigine or valproic acid, and non-users.

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Table 1. Participant characteristics, *n* (%) or mean (standard deviation)

	Before- and after-screening enrollees *n* = 7610	Before-screening enrollees *n* = 4516	After-screening enrollees *n* = 3094
Maternal race/ethnicity			
White	6605 (86.8%)	3969 (87.9%)	2636 (85.2%)
Hispanic	380 (5.0%)	206 (4.6%)	174 (5.6%)
Black	229 (3.0%)	119 (2.6%)	110 (3.6%)
Mixed	166 (2.2%)	92 (2.0%)	74 (2.4%)
Asian	135 (1.8%)	78 (1.7%)	57 (1.8%)
Others	93 (1.2%)	51 (1.1%)	42 (1.4%)
Maternal education			
Grade 12 or less	951 (12.5%)	549 (12.2%)	402 (13.0%)
Incomplete college	1354 (17.8%)	827 (18.3%)	527 (17.0%)
Completed college	2160 (28.4%)	1358 (30.1%)	802 (25.9%)
Post-college education	1381 (18.1%)	808 (17.9%)	573 (18.5%)
Married at enrollment	4811 (63.2%)	2966 (65.7%)	1845 (59.6%)
Maternal age at conception (years)	30.46 (5.3)	30.36 (5.1)	30.62 (5.7)
Folic acid supplementation	5219 (68.6%)	3156 (69.9%)	2063 (66.7%)
Alcohol consumption in first trimester			
None	5668 (74.5%)	3330 (73.7%)	2338 (75.6%)
Less than five drinks per week	1638 (21.5%)	1001 (22.2%)	637 (20.6%)
Five drinks per week or more	161 (2.1%)	100 (2.2%)	61 (2.0%)
Yes, quantity unknown	125 (1.6%)	73 (1.6%)	52 (1.7%)
Cigarette smoking in first trimester	1066 (14.0%)	617 (13.7%)	449 (14.5%)
Use of any antiepileptic drug*	7029 (92.4%)	4304 (95.3%)	2725 (88.1%)
Gestational age at birth (weeks)	39.00 (2.1)	38.94 (2.2)	39.08 (1.9)

Race/ethnicity was missing for two participants, maternal education for 1764, marital status at enrollment for 1773, folic acid supplementation for 108, alcohol consumption in the first trimester for 18, cigarette smoking in the first trimester for 12, and gestational age at birth for 75.

*Antiepileptic drugs included acetazolamide, alprazolam, carbamazepine, clobazam, clonazepam, clorazepate, diazepam, ethosuximide, felbamate, gabapentin, lamotrigine, levetiracetam, lorazepam, mephenytoin, oxazepam, oxcarbazepine, phenobarbital, phenytoin, primidone, tiagabine, topiramate, valproic acid, vigabatrin, zonisamide, or other. No enrollees reported first trimester use of ethotoin, methsuximide, phensuximide, paramethadione, or trimethadone.

Spontaneous abortion analyses. Subjects were followed up from enrollment or the beginning of gestational week 5 (whichever came last) until spontaneous abortion, elective termination, or the end of gestational week 20, whichever came first. Pregnancies that ended in a delivery beyond the end of gestational week 20 (the vast majority of pregnancies) had their follow-up truncated at the end of gestational week 20. We thus took into account gestational age at enrollment[10] and at the end of follow-up for each pregnancy. To explore variation in the incidence rate of spontaneous abortion by gestational age, we calculated the incidence rate in each gestational month as a fraction. The denominator was the number of person-months accrued by the cohort each gestational month, and the numerator was the number of spontaneous abortions observed during each gestational month.[19,20] Incidence rates in each gestational month were estimated for users and non-users of antiepileptic drugs, and the monthly incidence rate ratios were tested for homogeneity prior to pooling.

To evaluate the impact of gestational age at enrollment on the association of antiepileptic drug use versus.no use with spontaneous abortion, we compared relative risk estimates using three methods: (i) unconditional exact logistic regression models (not accounting for left truncation); (ii) exact logistic regression models conditional on gestational age at enrollment scaled in trimester, month, or week (see Supplementary Information for code details); and (iii) the summary incidence rate ratio with Mantel–Haenszel weights.

Major congenital malformation analyses. To assess variation in the risk of major congenital malformations by gestational age at enrollment, we estimated risk as a proportion where the denominator was the number of participants enrolled each gestational bimester (i.e., 2-month periods) and the numerator was the number of congenital malformations confirmed in those participants at any time during follow-up. Gestational age was aggregated in 2-month bins because of sparse data for non-users. We tested whether there was variation in the risk of major congenital malformations by gestational age at enrollment and whether it differed for antiepileptic-drug users versus non-users. To do this, we fitted a logistic regression model for major congenital malformations as a function of gestational age at enrollment, use of any antiepileptic drug versus no use, and a cross-product

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term between the two. Gestational age at enrollment was measured in 2-month intervals: enrollment in months 1 or 2 was coded as 1, in months 3 or 4 as 2, and so forth. We then conducted a Wald test on the cross-product term.

To evaluate the impact of gestational age at enrollment on estimates of the associations of congenital malformations and first trimester antiepileptic drug use (any antiepileptic drug use vs. no use, valproic acid monotherapy or combination therapy vs. no antiepileptic drug use, and valproic acid monotherapy vs. lamotrigine monotherapy), we compared the relative risk estimates with unconditional exact logistic regression models and exact logistic regression models conditional on gestational age at enrollment scaled in trimester, month, or week.

Stratified analysis: before-screening enrollment. Analyses were first conducted among all enrollees and then repeated among before-screening enrollees only. Analyses restricted to after-screening enrollees were not conducted because of the scarcity of outcomes in the unexposed: no spontaneous abortions and 3 cases of major congenital malformations.

Missing dates. The date of, or gestational age at, spontaneous abortion was missing in 119 (33%) subjects. These dates are needed to calculate incidence rates and rate ratios. Excluding subjects with missing dates may cause bias, as missingness was likely not at random. Further, information from one third of the cases would have been lost. Therefore, we employed several methods to handle missing values, as follows. To impute missing values in the main analysis, we sampled with replacement from among the observed gestational ages at spontaneous abortion. If the sampled gestational age was earlier than enrollment, we resampled until an appropriate value was found. Because follow-up is stopped at elective termination, we proceeded similarly with the 12 missing dates of elective termination (28%). More details can be found in the Supplementary Information.

Sensitivity analysis. To evaluate the sensitivity of the results to our handling of missing data, we first conducted a complete case analysis, where we excluded all spontaneous abortions and elective terminations with missing event date or gestational age at the event. As an alternative, we also used multiple imputations to create five complete data sets based on observed information. Details of methods and results are provided in the Supplementary Information.

All analyses were conducted with SAS 9.2 (SAS Institute Inc., Cary, NC, USA) except for the analysis of rates, which were conducted on Episheet 2008.[21]

RESULTS

Characteristics of the study population. Of the 8629 enrollees from the inception of the registry until March 2013, 593 women were lost to follow-up, 175 had unknown birth outcomes, 169 withdrew, 75 had unknown before/after-screening status, 4 enrolled after the end of pregnancy, and 3 had unknown gestational age at enrollment. These women were excluded from the analysis. The study population comprised 7610 participants. Table 1 shows participant characteristics. A total of 7029 women received antiepileptic drugs in the first trimester of gestation as monotherapy or combination therapy. Of these, 2433 women received lamotrigine, and 540 received valproic acid; 4516 participants (59%) were before-screening enrollees.

Gestational age at enrollment by exposure status. Enrollment spanned gestational weeks 1 to 42 (Figure 1). Median gestational age at enrollment (25–75th percentile) in users of any antiepileptic drug was 14 (8–22) weeks. In lamotrigine users, it was 13 (8–21); in valproic acid users, 14.5 (9–23); in non-users, 21 (14–28). Among before-screening enrollees, median gestational age at enrollment in users of any antiepileptic drug was 9 (7–13) weeks; it was 9 (6–13) in lamotrigine users, 10 (7–13) in valproic acid users, and 12 (9–16) in non-users.

Spontaneous abortion analyses. With 359 spontaneous abortions overall, the rate of spontaneous abortion among antiepileptic drug users declined from 115 to 10 events per 1000 person-months between gestational months 1 and 5 (Table 2). Among non-users, rates were lower and also decreased over the course of pregnancy, from 47 to 0 events per 1000 person-months between gestational months 2 and 5. Table 2 also shows the dynamic size of the study population, with 17.6 observed person-months from women under follow-up during their first month of pregnancy and 3983.8 person-months from women under follow-up during their fifth month of pregnancy. The relative risk for antiepileptic drug users compared with non-users was 5.1 (2.3–14.1) based on unconditional logistic regression analysis (Table 3). Conditioning on gestational week at

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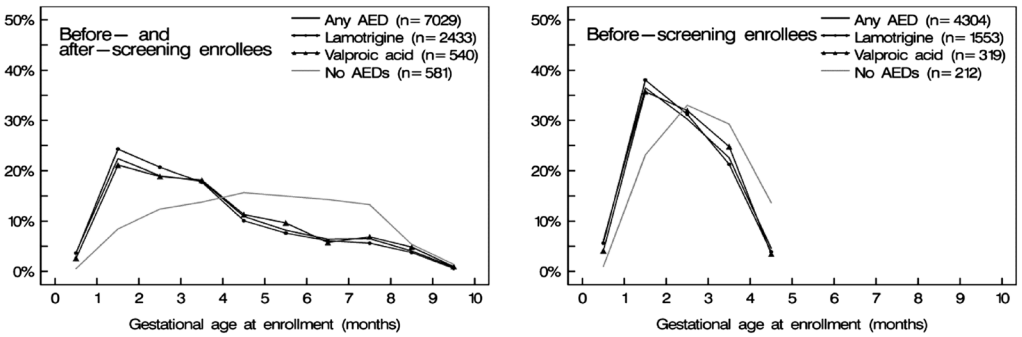

Figure 1. Distribution of gestational age at enrollment by antiepileptic drug use. AED, antiepileptic drug

Table 2. Incidence rate per 1000 person-months, and incidence rate ratio, of spontaneous abortion in gestational months 1 to 5

		Month 1	Month 2	Month 3	Month 4	Month 5	Pooled IRR
Before- and after-screening enrollees							
AED users	Events	2	97	163	54	37	
	Person-months	17.4	954.7	2374.7	3526.3	3779.9	
	Incidence rate	114.7	101.6	68.6	15.3	9.8	
Non-users	Events	0	1	3	2	8	
	Person-months	0.2	21.1	85.2	158.5	203.9	
	Incidence rate	0	47.4	35.2	12.6	0	
	Incidence rate ratio	—	2.1	2.0	1.2	—	2.1
	(95% confidence interval)		(0.3–15.4)	(0.6–6.1)	(0.3–5.0)		(0.9–4.6)
Before-screening enrollees							
AED users	Events	2	97	163	53	34	
	Person-months	17.4	952.2	3,368.7	3,336.9		
	Incidence rate	101.9	69.1	15.7	10.2		
Non-users	Events	0	1	3	2	0	
	Person-months	0.1	20.1	83.8	146.5	167.83	
	Incidence rate	0	49.8	35.8	13.7	0	
	Incidence rate ratio	—	2.1	1.9	1.2	—	2.0
	(95% confidence interval)		(0.3–14.7)	(0.6–6.1)	(0.3-4.7)		(0.9–4.4)

Incidence rates are events per 1000 person-months. Because of the small number of events among non-users, we report exact 95% confidence intervals. AED, antiepileptic drug; IRR, incidence rate ratio.

enrollment, the relative risk was 2.0 (0.9–5.6). The summary incidence rate ratio was 2.1 (0.9–4.6; homogeneity of the monthly incidence rate ratios was not rejected with a p-value = 0.76).

Of the abortions, 355 were reported by before-screening enrollees. Incidence rates in this group were very similar to those in all enrollees combined. Before-screening enrollees had a lower unconditional relative risk, 3.1 (1.4–8.5), than all enrollees, 5.1. However, when conditioned on gestational age in weeks, or when incorporating person-time accrued by each study subject in the incidence rate ratio, point estimates did not substantially differ from those of all enrollees.

Major congenital malformation analyses. After exclusion of pregnancies that ended in spontaneous abortion, 7251 enrollees were eligible for analyses of major congenital malformations; 216 fetuses or infants with major malformations were identified. Antiepileptic drug users enrolled in the first 2 months of pregnancy had a risk of major congenital malformations of 29 cases per 1000 pregnancies; the risk increased in those who enrolled later (Figure 2). In non-users, the risk was 21 per 1000 among those who enrolled in the first 2 months of pregnancy, and the risk decreased in those who enrolled later. Confidence intervals were wide because of the small number of cases. The trend divergence between users and non-users was not statistically significant ($p = 0.17$). The relative risk of major congenital malformations in antiepileptic drug users compared with non-users was 3.1 (1.4–8.5; Table 3) in unconditional analyses; it was not substantially altered by conditioning on gestational age at

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Table 3. Relative risk of spontaneous abortion and congenital malformations

		Before- and after-screening enrollees					Before-screening enrollees				
		Exposed		Reference group		Relative risk (95% confidence interval)	Exposed		Reference group		Relative risk (95% confidence interval)
		Events	*n*	Events	*n*		Events	*n*	Events	*n*	
Any AED versus no AED and spontaneous abortions											
Unconditional logistic regression		353	7029	6	581	5.1 (2.3–14.1)	349	4304	6	212	3.1 (1.4–8.5)
Conditional on: trimester of enrollment						2.7 (1.2–7.6)					2.5 (1.1–7.1)
	Month of enrollment					2.2 (1.0–6.2)					2.1 (0.9–5.9)
	Week of enrollment					2.0 (0.9–5.6)					1.9 (0.8–5.4)
Summary incidence rate ratio						2.1 (0.9–4.6)					2.0 (0.9–4.4)
Any AED versus no AED and major congenital malformations											
Unconditional logistic regression		210	6676	6	575	3.1 (1.4–8.5)	117	3955	3	206	2.1 (0.7–10.2)
Conditional on: trimester of enrollment						3.2 (1.4–9.0)					2.1 (0.7–10.4)
	Month of enrollment					3.2 (1.4–9.0)					2.1 (0.7–10.3)
	Week of enrollment					3.2 (1.4–9.0)					2.0 (0.7–10.1)
Valproic acid versus no AED and major congenital malformations											
Unconditional logistic regression		47	505	6	575	9.7 (4.1–28.1)	27	285	3	206	7.1 (2.1–36.9)
Conditional on: trimester of enrollment						9.8 (4.1–28.4)					7.3 (2.2–38.4)
	Month of enrollment					9.5 (4.0–27.8)					7.0 (2.1–36.8)
	Week of enrollment					9.5 (3.9–27.9)					6.9 (2.0–36.3)
Valproic acid versus lamotrigine in monotherapy and major congenital malformations											
Unconditional logistic regression		29	322	34	1723	4.9 (2.8–8.4)	15	177	22	1069	4.4 (2.1–9.1)
Conditional on: trimester of enrollment						4.9 (2.9–8.5)					4.4 (2.1–9.1)
	Month of enrollment					5.0 (2.9–8.6)					4.5 (2.1–9.3)
	Week of enrollment					5.1 (2.9–8.9)					4.5 (2.1–9.4)

All confidence intervals from logistic regression models are exact confidence intervals.
AED, antiepileptic drug.

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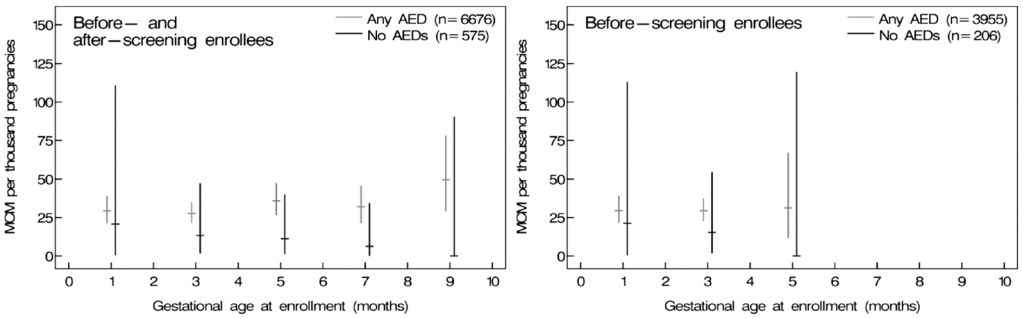

Figure 2. Risk of major congenital malformations by gestational age at enrollment among users and non-users of antiepileptic drugs, in 2-month bins. *Note*: Antiepileptic drug use refers to first-trimester use. Vertical lines represent confidence intervals. AED, antiepileptic drug

enrollment in trimester, month, or week. Unconditional and conditional analyses were also similar in comparisons of valproic acid users with non-users of any antiepileptic drug (relative risks 9.5–9.8) and in comparison of valproic acid monotherapy and lamotrigine monotherapy (relative risks around 5).

Among before-screening enrollees, there were 120 fetuses or infants with congenital malformations. In antiepileptic drug users, the risk was stable around 30 cases per 1000 pregnancies across all gestational ages at enrollment (Figure 2); in non-users, it decreased from 21 per 1000 to 0, with wide overlapping confidence intervals. Relative risks were lower in before-screening enrollees than among all enrollees in all comparisons (Table 3). Results did not change when conditioning by gestational age at enrollment. The comparison between valproic acid monotherapy and lamotrigine monotherapy gave the smallest change in estimated effect when restricting to before-screening enrollees.

Sensitivity analyses. In the complete case analysis, we excluded 131 pregnancies: 119 due to missing date at spontaneous abortion and 12 due to missing date at elective termination; 126 were before-screening pregnancies, and 129 were antiepileptic drug users. In the remaining 7479 pregnancies, incidence rates were lower, as expected (Supplementary Table 1), and relative risks remained practically unchanged. Likewise, in the multiple imputation analysis, incidence rates in all pregnancies combined and in before-screening pregnancies only were similar to the main analysis (see Supplementary Information).

DISCUSSION

Enrollment in the North American AED Pregnancy Registry is spread over a broad range of gestational ages; antiepileptic drug users tend to enroll earlier than non-users. For spontaneous abortion, analyses that conditioned on gestational month or week at enrollment resulted in risk ratios that were similar to the incidence rate ratio. The risk of major congenital malformations was overestimated in analyses that incorporated after-screening enrollees. The comparison of users of two active treatments was less sensitive to conditioning or restricting than comparisons of drug users to non-users, because users of different antiepileptic drugs were more similar in terms of gestational age and before/after prenatal screening status at enrollment.

The main limitation of this study is that gestational age at spontaneous abortion or elective termination was missing for some subjects; the registry was not designed to evaluate this outcome. However, our estimate of the risk of spontaneous abortion among unexposed early enrollees was consistent with US estimates,[22,23] and our rates were in the range reported for other cohorts.[20] Our results were robust in sensitivity analyses related to the treatment of the missing information. It is unlikely that women reported gestational age incorrectly, because these women are in close contact with the healthcare system (one would expect that they are knowledgeable about their gestational age), volunteered to provide information on their pregnancies (thus likely to report this information correctly), and were interviewed soon after the event (thus likely to recall accurately). If gestational age had been reported randomly around the true value by exposed and unexposed, we would not expect the point estimates to be affected. As we focus on some methodological aspects, we present only unadjusted results. Although having a group of unexposed women whose information can be used for comparisons within the registry is extremely valuable, the relative small size of the unexposed group resulted in imprecise estimates.

Pregnancy exposure registries are encouraged to enroll unexposed women to allow internal comparisons.[7,13] In this registry, antiepileptic drug users enrolled earlier in gestation than non-users, as found in other pregnancy

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cohorts.[24] The difference between comparison groups in the distribution of gestational age at enrollment may be less apparent in cohorts derived from automated data but may be nevertheless present.[25] Comparison groups in research on early-pregnancy events (e.g., spontaneous abortion) should have similar distributions of gestational age at enrollment (or start of follow-up), and all subjects should enroll in early pregnancy. Consider a scenario where the exposed subjects enroll earlier than the unexposed in a study of spontaneous abortion or other early-pregnancy events, and the true association is null. Because the earliest events in unexposed subjects are missed, analyses that ignore gestational age at enrollment can find a spurious association. Even when a real association exists, relative risks may be overestimated. This can be understood in the context of immortal person-time bias[26]: later enrollees contribute less time at risk for spontaneous abortion or none if they enroll after gestational week 20. Further, as the risk decreases over gestation, person-time contributed by later enrollees carries a lower risk of the outcome. As an example, in our study, only three spontaneous abortions occurred in women who enrolled after prenatal screening. With left truncation, when mean gestational age at start of follow-up in a cohort study differs by over 6–10 days across exposure levels, unconditional analyses of spontaneous abortions may result in bias of over 20% in estimates of relative risks.[8] A comparable bias has been reported for time to pregnancy with differential left truncation.[27]

A simulation study on spontaneous abortions showed that unconditional analyses, which do not account for left truncation, may estimate relative risks biased by over 20% when the mean gestational age at beginning of follow-up differs by over 6–10 days across exposure levels in different scenarios.[8] The authors recommend Cox regression to handle this left truncation. Cox models have also been recommended for studies on time-dependent exposures and spontaneous abortion[10] and other cases of left truncation.[28] We have used logistic regression conditional on gestational age at enrollment and a Mantel–Haenszel-weighted summary incidence rate ratio. The partial and conditional likelihood functions to estimate the regression coefficients in Cox and conditional logistic regression, respectively, have a common structure.[29] Therefore, estimated hazard and odds ratios derived from the same datasets will be similar if the time scale chosen to form the risk sets is the same. To show this, we ran a Cox regression model with gestational age at enrollment scaled in weeks among before-screening enrollees to estimate the risk of spontaneous abortions related to use of any antiepileptic drug versus non-use. The hazard ratio was 1.9 (0.8–4.2), to compare with 2.0 (0.9–4.4) from Table 3. The Cox model required gestational age at spontaneous abortion be known, for which we used imputed values, whereas this information was not needed in the conditional logistic regression analysis. Cox regression can accommodate time-varying exposures, which are preferred, when there is risk for immortal time bias, to the time-unvarying exposure used with logistic regression.[30] In our spontaneous abortion analyses, there is a risk for misclassified immortal time if women started antiepileptic drug treatment at some point during the first trimester of pregnancy and are treated as exposed during the entire period. Out of 7029 women, 158 (2.3%) were in this situation; therefore, their misclassified time at risk of spontaneous abortion (from the latest of gestational week 5 or enrollment to the moment when they started treatment) is very small and unlikely to alter relative risk estimates.

For studies on congenital malformations or other outcomes that are routinely assessed with prenatal screening, enrollment must happen prior to prenatal screening, and as early as possible. This is because not only prenatal screening results but also several other signs can hint at possible complications. Early enrollment is also needed to capture fetal malformations in electively terminated pregnancies, because elective terminations generally occur relatively early in pregnancy. In our study, 50% of them took place before gestational week 17. Trends in the risk of major congenital malformations by gestational age at enrollment initially appeared to differ by exposure status; the later antiepileptic drug users enrolled, the higher their risk for congenital malformations. This trend was driven by after-screening enrollees. The potential for selection bias in exposure pregnancy registries has been widely discussed.[3,5,7,13,31] However, the direction of the bias is not necessarily clear. Underestimation of risk can be expected if women with normal prenatal screening results are more likely to enroll than women who are aware of pregnancy complications or undesired outcomes. Overestimation of the risk is to be expected when enrollment is more likely after receiving abnormal test results. Artificial associations may arise if these behaviors are differentially distributed among exposure levels.[4] In this study, with a median gestational age at enrollment of 9 weeks among before-screening enrollees and 26 among after-screening enrollees, we show that including after-screening enrollees in the analyses probably biases the relative risk estimates away from the null when comparing antiepileptic drug use with no use. The comparison of valproic acid and lamotrigine was less affected by gestational age at enrollment and by the inclusion of after-screening enrollees. This evidence supports the use of

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comparative effectiveness and safety approaches (i.e., comparing users of different drugs) in pregnancy registries to minimize selection bias due to differential time of enrollment. In addition, users of different drugs for the same indication (e.g., epilepsy) will tend to be more comparable, thus reducing confounding by indication.

In conclusion, our empirical results support recommendations to condition on gestational age at enrollment, or to estimate incidence ratios per gestational month, in research on spontaneous abortion in pregnancy exposure registries to account for left truncation and dynamic sample size. In studies of major congenital malformations, restriction to before-screening enrollees appears to be vital to avoid selection bias. Comparisons of active treatments seem to be more robust, because users of different drugs tend to have similar distributions of gestational age at enrollment. We endorse efforts to ensure early enrollment of all participants, exposed and unexposed.

CONFLICT OF INTEREST

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KEY POINTS

- Gestational age at enrollment may differ between treated and untreated groups in studies of drug safety in pregnancy, such as pregnancy exposure registries.
- In studying early-pregnancy events (e.g., spontaneous abortions), it is key to ensure that all treatment groups have comparable time at risk for the outcome and that left truncation is handled correctly in the analysis.
- Comparison of active treatments may be less vulnerable to these issues than the comparison of treated and untreated groups.
- In studying outcomes identified through prenatal testing, enrollment should precede the tests.

ETHICS STATEMENT

The study has been approved annually by the Human Studies Committee of the Massachusetts General Hospital and Partners HealthCare.

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