January 2020

Live Birth Bias In Epidemiological Study Of Timing Specific Exposure Effect During Pregnancy And Child Health: A Simulation Study

Ngoc Tram Nguyen
nguyentram.christine@gmail.com

Live birth bias in epidemiological study of timing specific exposure effect during pregnancy and child health: a simulation study

by

Ngoc Tram Nguyen

A thesis in partial satisfaction of the requirements for the degree of

Master of Public Health

in

Chronic Disease Epidemiology

Thesis Mentor: Zeyan Liew, PhD, MPH

Yale School of Public Health

Spring 2020

ABSTRACT

In reproductive and perinatal epidemiological studies, measurement of child health outcomes that can only be ascertained in live born children may be incomplete since only 60 – 70% of fertilized eggs result in live births and early pregnancy loss is often undetected. Studies assessing outcomes among live born children are subject to live birth bias, a phenomenon previously proposed as a form of collider-bias in which conditioning on live-birth status induces a non-causal association between exposure and outcome. In this study, we expanded a previously proposed common structure of this bias to evaluate its impact on the estimation of time-specific prenatal exposure effects on child health outcome, using causal diagrams. We used Monte Carlo simulation techniques to investigate two scenarios in which prenatal exposures led to pregnancy loss. Our findings confirmed previous simulation findings showing biased estimates of prenatal exposure effects on child outcome risk, assuming a true null association between each exposure and the outcome and using trimesters to characterize the exposure timing. We observed larger bias sizes when the effect size of the exposure-fetal survival relations increased and/or when other unmeasured and uncontrolled risk factors had stronger effect on both fetal survival and the outcome. Our study underlines the needs for the development of analytic methods that adjust for live birth bias in scenarios accounting for time-specific exposure effects and time-specific selections.

Keywords: live-birth bias, quantitative bias analyses, perinatal epidemiology, pregnancy cohort, time varying exposures, causal diagrams

ACKNOWLEDGMENTS

A special thanks to Dr. Zeyan Liew, PhD, MPH who mentored the candidate and contributed significantly to the conception and analysis of the study as well as the revision of the manuscript.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

BACKGROUND

Cohort studies examining the effect of pregnancy exposures on child health outcomes are susceptible to live birth bias, a type of selection bias common in perinatal epidemiology.[1] A host of factors, including those that are genetic, hormonal, immunological, environmental, and lifestyle-related, can lead to early pregnancy loss, spontaneous abortions, and miscarriages, leading to only an estimate of about less than 60-70% of all conceptions surviving to birth.[1,2,3,4] The lost fetuses are not accounted for in risk estimation when studies restrict samples to only live births, thus resulting in biased estimates of exposure effects on outcomes of interest.[1,5] When studying long-term health outcomes that can only be ascertained after birth and during childhood, only live born infants are qualified candidates to be in the at-risk population for these disease outcomes, making the restriction of study subjects to only live born children inevitable.[1]

Several quantitative bias analyses using simulations have investigated biased estimates due to live birth bias, including one study assessing the effect of prescriptive drug use during pregnancy on preeclampsia[6], one assessing the effect of maternal smoking on preeclampsia at delivery,[7] and another assessing the impact of prenatal exposure to perfluoroalkyl substances (PFAS) effect on neurodevelopmental outcome such as attention-deficit/hypersensitivity disorder (ADHD) in young children.[1] Pregnancy cohort studies face numerous challenges in obtaining accurate numbers of pregnancy losses especially in early gestations, and that maternal and child health outcomes of interests might only be ascertainable among live births.[8] Simulation studies, therefore, provide an opportunity to study the structure and magnitude of live birth bias.

Pregnancy loss at different time periods during pregnancy have been studied and documented. The incidence of fetal loss after implantation and before clinical detection was

estimated to range from 20-30% by studies using biomarkers such as urinary concentrations of human chorionic gonadotropin to detect pregnancy loss.[9,10] Fetal loss rate decreases with increasing gestational age; most pregnancy losses occur before week 12 (about 80% of all pregnancy loss cases occur within the first trimester) and drops substantially afterwards.[9,11,12] Certain environmental and lifestyle risk factors can impact fertility and fetal survival at certain critical windows of susceptibility. For instance, smoking has been found to affect chances to conceive as well as miscarriage, stillbirth, and infant mortality during the first 12 months of life.[13,14] Bisphenol A (BPA) and phthalates have also been associated with the highest rate of miscarriage detected at or before week 20 or 22.[15,16,17,18] While these "critical risk windows" are estimates and not definite, studies looking at the differential effects of prenatal exposures on child outcome at different times in pregnancy often use trimesters to define these time windows.

While existing studies have quantified live birth bias for the association of overall or average exposure throughout during pregnancy and child health, no study has yet evaluated the timing-specific nature of exposures and selection bias due to pregnancy loss throughout gestations. In this study, we built on the previously proposed structure of the live birth bias by accounting for timing specific exposures, using directed acyclic graphs (DAGs) to illustrate two scenarios in which conditioning on live births can bias the estimation of pregnancy timing exposure effects on the outcome, especially when the exposures at each time could affect fetal survival or pregnancy loss but do not causally affect the outcome.[1] We hypothesized that time-specific exposure effects on child health outcome will be influenced by live birth bias when exposures affect pregnancy loss and analyses are restricted to live births only. The direction and magnitude of such bias would depend on the structure of the bias, including whether the exposures vary across the pregnancy

period and how strongly the exposures affect fetal survival or pregnancy loss at each timing in the presence of unmeasured causal determinants of fetal loss and the outcome.

METHODS

Directed acyclic graph

A DAG is a graphical model with pathways representing the relations between variables of interest, consisting of nodes (variables) connected by one-directional arrows with directed edges in a way that creates no closed loop paths.[19] A causal path is a directed path going along the arrows, leading to direct and indirect effect between an exposure and outcome of interest. A backdoor path, on the other hand, illustrates a classic example of confounding bias in which a third variable, a confounder, causes the exposure and outcome. Controlling for this third variable closes the backdoor path, removing the bias (Figure S1, Supplemental). A DAG representing collider bias, another major source of bias in epidemiology, includes arrows pointing towards a single variable, a collider, from other variables. Conditioning on a collider will open the otherwise closed collider path, inducing a non-causal association between the exposure and outcome (Figure S1, Supplemental). Current literature has shown that survivor bias, loss to follow-up, and non-response bias are forms of collider bias.[20,21]

Structure of live birth bias

Live birth bias has been illustrated using DAG as an example of collider bias or competing risk bias common in epidemiologic studies, particularly perinatal studies examining the effects of prenatal exposures to environmental influences on long-term health neurodevelopmental outcomes in the offspring (Figure 1).[1] These outcome can only be ascertained among live-born children, under the assumption of no loss to follow up between birth and the time of exposure measurement. If the exposure contributes to pregnancy loss in the presence of other uncontrolled common causes of fetal loss and the outcome, restricting the source population to or conditioning on live births opens the collider path, leading to collider bias.[1]

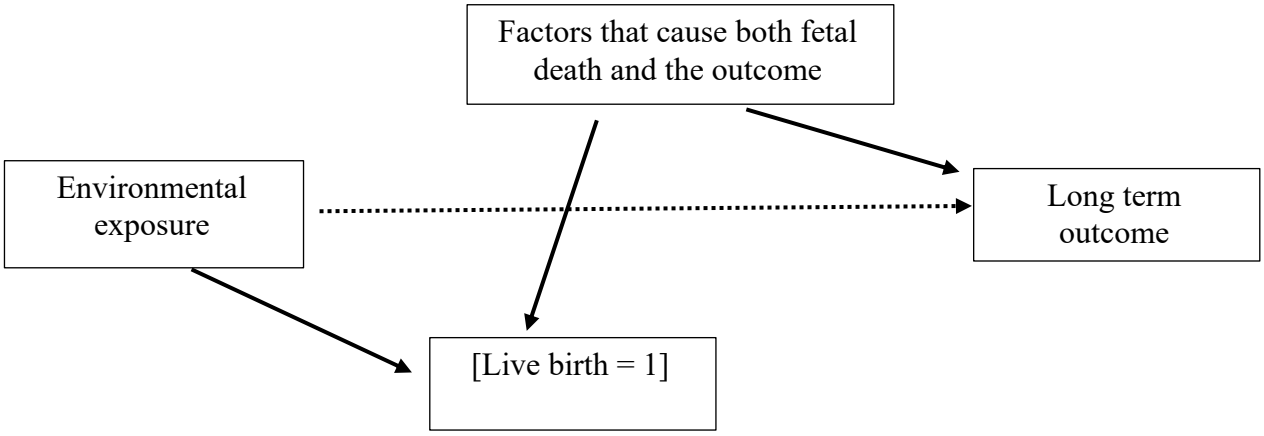

Figure 1. Basic structure of live birth bias in pregnancy cohort studies looking at prenatal exposure to environmental factors on birth outcomes and long-term health outcomes in children proposed.[1] Conditioning on live born fetuses will open a collider path from environmental exposure to outcome via unmeasured unknown common causes of fetal survival and the outcome.

Given the complex etiology of early pregnancy loss and miscarriage, those who were exposed to the risk factor of interest but survived might be less likely to have other harmful factors. Among the surviving population, the exposed subjects have lower prevalence of other factors that affect fetal survival, and if these factors are also risk factors for the outcome, we might find a spurious association suggesting a lower risk for outcome among the exposed.

Consideration of exposure timings in pregnancy

Pregnancy trimesters are commonly used as a proxy of time to characterize milestone events of fetal development and measure prenatal exposures, fetal growth events, and fetal loss events for clinical and research purposes. Reproductive and perinatal epidemiological studies often report pregnancy-trimester effects for the exposure and the maternal and child health outcomes of interest. Trimester specific clinical guidelines have been developed to help detect early signs of adverse infant outcomes or prevent adverse maternal outcomes; for instance, guidelines and recommendations for aspirin use to prevent preeclampsia have focused on second and third trimesters.[22] Nevertheless, trimesters are merely proxy measures as these risk windows may differ by the intensity and mechanism of specific exposures and outcomes.[23] We used trimesters in this study to conceptualize exposure time windows while assuming that these timings could be applied to any three time points during pregnancy.

Simulation scenarios

We used DAGs to illustrate the structural relations of trimester specific exposures (main independent variables), fetal survival at each trimester, child outcome (main dependent variable), and unmeasured or unknown common causes of fetal loss and the outcome (Figures 2 – 3). Our simulations assessed the impact of live birth bias induced by conditioning on selection (fetal survival or chance of pregnancy loss) at first trimester (S1=1), second trimester (S2=1), and third trimester (S3=1) to estimate the effects first trimester exposure (E1), second trimester exposure (E2), and third trimester exposure (E3) on a disease risk in the offspring in childhood (D). Fetal survival at each trimester was assumed to be affected by each trimester-specific exposure and a set of unmeasured or unknown time-invariant risk factors (U) over the course of pregnancy, including

genetic predisposition or any pre-existing maternal condition. Two scenarios were created from the basic structure presented in Figure 1, assuming that each of the trimester exposure variables (E1 – E3) are independent or dependent, that their causal effects on fetal survival at each trimester is short term, and that none of them caused the outcome. Our simulation reflected the reality where fetuses surviving at a later trimester must have survived through the preceding trimester(s), hence conditioning on S3=1would inevitably condition on S1=1 and S2=1.

In Scenario 1 (Figure 2), we assumed pregnancy trimester-specific exposures are marginally independent and encompassing factors that occur only once in pregnancy but may have considerable impact on fetal survival. Examples of these type of exposures include infection to a certain agent (e.g. chlamydial infection), sudden major injuries, or sudden stressful life events (e.g. death of relatives).[24,25,26]

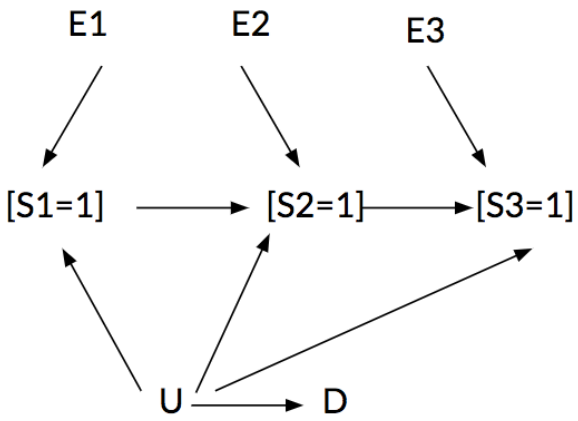

Figure 2. A directed acyclic graph (DAG) that illustrates the bias structure in scenario 1. The trimester-specific exposures (E1 to E3) are marginally independent. Each trimester-specific exposure variable (E1 to E3) affects the selection node at a relevant time (S1 to S3). Conditioning on these selection nodes induce open collider paths from the exposures to the outcome via a set of unmeasured or unknown risk factors (U) of the outcome that also affect selections.

In Scenario 2 (Figure 3), we assumed marginal dependence between the exposures such that prior exposures affect the subsequent exposures later in pregnancy. Exposures with

moderate to strong correlations may include persistent maternal factors or lifestyle habits (e.g. smoking or caffeine intake) or persistent environmental chemicals that have a long biological half-life (e.g. PFAS).[27,28]

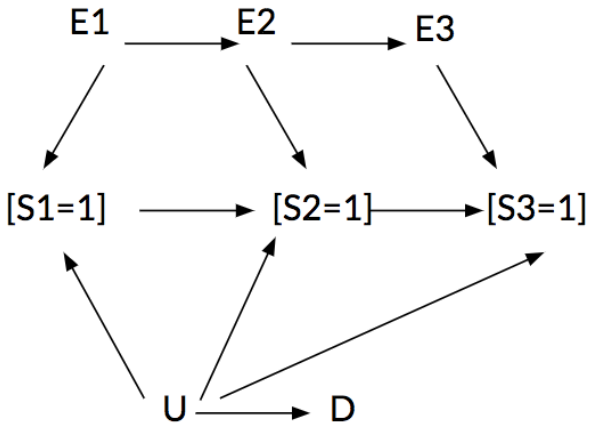

Figure 3. A directed acyclic graph (DAG) that illustrates the bias structure in scenario 2. The trimester-specific exposure (E1 to E3) are marginally dependent. Each trimester-specific exposure variable affects the selection node at the relevant time (S1 to S3). Conditioning on these selection nodes induce open collider paths from the exposures to the outcome via a set of unmeasured or unknown risk factors (U) of the outcome that also affect selections.

Simulations and statistical analysis

We conducted all analyses in SAS 9.4 (SAS Institute, Cary, NC, USA), using Monte Carlo techniques to perform simulations on a hypothetical cohort of 100,000 conceptions, a size that resembles that of the Danish National Birth Cohort (DNBC).[1,29] Of the total numbers of conceptions, 85% survived at the first trimester, 90% the surviving fetuses survived at the second trimester, and 95% of the remaining fetuses resulted in live births. At baseline, we simulated ~70% of all conceptions were born alive among those unexposed to the trimester specific exposures (E1-E3) and those without the risk factor U present. We assumed the prevalence of each trimester-specific exposure to be 25% in scenario 1 and increase at second and third trimesters due to the dependence of the exposure variables in scenario 2. A range of priors was assigned to the effect size of the associations between U and the outcome (OR_{U-D}), U and timing-specific selections

(OR_{U-S1}, OR_{U-S2}, OR_{U-S3}), and trimester specific exposures and fetal survival (OR_{E1-S1}, OR_{E2-S2}, OR_{E3-S3}). The disease prevalence was kept under 10% for the odds ratios to approximate relative incidence risk ratios in cohort studies. Tables 1 and 2 provide details of these priors and formula used to simulate these variables.

We assumed that each trimester exposures do not cause the outcome in both scenarios and conducted logistic regression analysis in each simulated dataset under different sets of priors to estimate the odds ratios for each timing exposures on the outcome, conditioning on live births only (S3=1), and evaluated how estimates deviated from the true effect for each exposure variable which was assumed to be null (OR=1.0). We computed 95% simulation intervals (SI), using 2.5, 50, and 97.5 percentiles, following 100 replications, for each simulation. For each dataset, we conducted two models: in model A each trimester-specific exposure variable was analyzed separately while in model B all three exposure variables were assessed simultaneously.

In sensitivity analyses, we tested the effect estimates when the strength of the associations between exposures and selections at all trimesters decreased from early to later gestations in Scenario 2, e.g. we assumed that the first trimester exposure (E1) has a stronger effect on selection (OR_{E1-S1}=0.15, 0.25), and the effect size gradually attenuated for exposure in the second trimester (OR_{E2-S2}=0.50, 0.70) and the third-trimester (OR_{E3-S3}=0.80, 1.00).

Table 1. Priors for the simulations

Variable [a]	Abbreviation	Prevalence	Specified relation (ORs) between variables
First trimester exposure	E1	25%	No causal determinant
Second trimester exposure	E2	25%	No causal determinant when exposures are uncorrelated. When exposures are marginally dependent, $OR_{(E1-E2)}$ = 8.0 to represent strong effects from prior to later exposures.
Third trimester exposure	E3	25%	No causal determinant when exposures are uncorrelated. When exposures are marginally dependent, $OR_{(E1-E2)}$ = 8.0 to represent strong effects from prior to later exposures.
Fetal survival at 1st trimester	S1=1	85% of all conceptions	OR_{E1-S1} = 0.70, 0.50, 0.30 or 0.10 in primary analysis OR_{E1-S1} = 0.15 or 0.25 in sensitivity analysis of scenario 2
Fetal survival at 2nd trimester	S2=1	90% of surviving fetuses	OR_{E2-S2} = 0.70, 0.50, 0.30 or 0.10 in primary analysis OR_{E2-S2} = 0.50 or 0.70 in sensitivity analysis of scenario 2
Fetal survival at 3rd trimester	S3=1	95% of surviving fetuses	OR_{E3-S3} = 0.70, 0.50, 0.30, or 0.10 in primary analysis OR_{E3-S3} = 0.80 or 1.00 in sensitivity analysis of scenario 2
Unknown and unmeasured risk factors	U	20% or 40%	$OR_{U-S1} = OR_{U-S2} = OR_{U-S3}$ = 0.20
Disease outcome	D	2 % - 8%	OR_{D-U} = 2.0, 5.0 or 10.0

[a] All variables were generated as binary variables (1 = yes, 0 = no)

Table 2. Formula for the simulated binary variables in scenarios 1 and 2

Variable [a]	Scenario 1	Scenario 2
U	U~B (1, 0.40)	U~B (1, 0.40)
D	• D~B (1, (1/ (1 + exp(-log(P(D1 = 1)/(1 – P(D1 = 1)) + log(OR_{D-U})*U))))	• D~B (1, (1/ (1 + exp(-log(P(D1 = 1)/(1 – P(D1 = 1)) + log(OR_{D-U})*U))))
E1	• E1~B (1, 0.25)	
E2	• E2~B (1, 0.25)	• E2~B (1, (1 / (1 + exp(-log(0.25/0.75) + log(OR_{E1-E2})*E1))))
E3	• E3~B (1, 0.25)	• E3~B (1, (1 / (1 + exp(-log(0.25/0.75) + log(OR_{E2-E3})*E2))))
S1	• S1~B (1, (1 / (1 + exp(-log(P(S1 = 1)/(1 – P(S1 = 1)) + log(OR_{E1-S1})*E1 + log(OR_{U-S1})*U))))	
S2	• S2~B (1, (1 / (1 + exp(-log(P(S2 = 1)/(1 – P(S2 = 1)) + log(OR_{E2-S2})*E1 + log(OR_{U-S2})*U)))) | S1 = 1	
S3	• S3~B (1, (1 / (1 + exp(-log(P(S3 = 1)/(1 – P(S3 = 1)) + log(OR_{E3-S3})*E1 + log(OR_{U-S3})*U)))) | S2 = 1	

[a] All variables were generated as binary variables (1 = yes, 0 = no)

RESULTS

In the first scenario where we assumed trimester specific exposures were marginally independent, a "protective" effect of each trimester-specific exposure on the outcome was observed among live births (S3=1) when the true causal effect is null. For each simulated dataset, the strength of these inverse associations observed were stronger in early trimester and they became slightly weaker or closer to null in later trimesters. The biased effect sizes became larger when the effect of exposures on selections and/or the effect size of the association between U and the outcome (OR_{U-D}) increased. The largest biased effect estimates observed were $OR_{E1-D} = 0.71$, $OR_{E2-D} = 0.71$, $OR_{E3-D} = 0.74$ in model A that analyzed each exposure variable separately, and the results were nearly identical between models A and B. These biased effect estimates moved closer to the null when the strength of association between U and selections decreased from 0.20 to 0.50 or when the prevalence of U decreased from 40% to 20% (Tables S1 and S2, Supplemental).

When exposures were marginally dependent in scenario 2, the observed effect size between the exposure and the outcome were even more biased away from the null compared to their counterparts in scenario 1 (Table 3). With strong associations between preceding exposures and the following exposures during pregnancy ($OR_{E1-E2} = OR_{E2-E3} = 8.0$), the largest observed biased effect estimates were $OR_{E1-D} = 0.63$, $OR_{E2-D} = 0.61$, $OR_{E3-D} = 0.68$ in model A. The bias size of the estimates in model A also decreased when the strength of the associations between the exposure decreased, e.g. $OR_{E1-E2} = OR_{E2-E3} = 1.50$ (Table S3, Supplemental).

Table 3. Odds ratios and 95% simulation intervals [a] of the associations observed between the trimester specific exposures (E1-E3) on the outcome (D) in scenario 1, conditioning on live-birth cohort (S3=1) status when exposures are marginally independent and the true causal effects of all exposure variables and the outcome are null (OR=1.0)

		$OR_{E1-S1} = OR_{E2-S2} = OR_{E3-S3} =$							
	OR_{U-D}	0.70		0.50		0.30		0.10	
		Model A	Model B	Model A	Model B	Model A	Model B	Model A	Model B
E1	2	0.98 (0.96 – 0.99)	0.98 (0.96 – 0.99)	0.96 (0.95 – 0.98)	0.96 (0.95 – 0.98)	0.94 (0.93 – 0.95)	0.94 (0.93 – 0.96)	0.93 (0.91 – 0.95)	0.93 (0.91 – 0.95)
E2		0.99 (0.98 – 1.00)	0.99 (0.98 – 1.00)	0.98 (0.96 – 0.99)	0.98 (0.96 – 0.99)	0.96 (0.94 – 0.98)	0.96 (0.94 – 0.98)	0.93 (0.91 – 0.94)	0.93 (0.91 – 0.95)
E3		1.01 (1.00 – 1.02)	1.01 (1.00 – 1.02)	1.00 (0.98 – 1.01)	1.00 (0.98 – 1.01)	0.97 (0.96 – 0.99)	0.97 (0.96 – 0.99)	0.94 (0.93 – 0.96)	0.94 (0.93 – 0.96)
E1	5	0.96 (0.95 – 0.97)	0.96 (0.95 – 0.97)	0.92 (0.91 – 0.93)	0.92 (0.91 – 0.93)	0.86 (0.85 – 0.87)	0.86 (0.85 – 0.87)	0.80 (0.79 – 0.82)	0.81 (0.79 – 0.82)
E2		0.96 (0.95 – 0.97)	0.96 (0.95 – 0.97)	0.93 (0.91 – 0.94)	0.93 (0.92 – 0.94)	0.88 (0.87 – 0.89)	0.88 (0.87 – 0.90)	0.80 (0.79 – 0.82)	0.80 (0.79 – 0.82)
E3		0.99 (0.98 – 1.00)	0.99 (0.98 – 1.00)	0.96 (0.95 – 0.98)	0.96 (0.95 – 0.98)	0.91 (0.90 – 0.93)	0.91 (0.91 – 0.93)	0.83 (0.81 – 0.84)	0.83 (0.82 – 0.84)
E1	10	0.94 (0.93 – 0.95)	0.94 (0.93 – 0.95)	0.89 (0.88 – 0.90)	0.89 (0.88 – 0.90)	0.80 (0.79 – 0.81)	0.80 (0.79 – 0.81)	0.71 (0.70 – 0.72)	0.71 (0.70 – 0.72)
E2		0.95 (0.94 – 0.96)	0.95 (0.94 – 0.96)	0.90 (0.89 – 0.91)	0.90 (0.89 – 0.91)	0.83 (0.82 – 0.84)	0.83 (0.82 – 0.84)	0.71 (0.70 – 0.72)	0.71 (0.70 – 0.72)
E3		0.98 (0.97 – 0.99)	0.98 (0.97 – 0.99)	0.94 (0.93 – 0.95)	0.94 (0.93 – 0.95)	0.87 (0.87 – 0.88)	0.87 (0.86 – 0.88)	0.74 (0.74 – 0.76)	0.75 (0.74 – 0.76)

[a]Logistic regression models were used to estimate each of the binary trimester specific exposure effect (E1-E3) on a binary outcome (D). Model A included each trimester exposure effect separately while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U-S1} = OR_{U-S2} = OR_{U-S3} = 0.20$, and the prevalence of U = 40%.

Table 4. Odds ratios and 95% simulation intervals [a] of the associations observed between the trimester specific exposures (E1-E3) on the outcome (D) in scenario 2, conditioning on live-birth cohort (S3=1) status when exposures are marginally dependent and the true causal effects of all exposure variables and the outcome are null (OR=1.0), given strong associations between trimester specific exposures ($OR_{E1-E2} = OR_{E2-E3} = 8.00$)

		$OR_{E1-S1} = OR_{E2-S2} = OR_{E3-S3} =$							
	OR_{U-D}	0.70		0.50		0.30		0.10	
		Model A	Model B	Model A	Model B	Model A	Model B	Model A	Model B
E1	2	0.97 (0.95 – 0.98)	0.97 (0.96 – 0.99)	0.95 (0.93 – 0.96)	0.96 (0.95 – 0.98)	0.92 (0.91 – 0.94)	0.94 (0.93 – 0.96)	0.92 (0.89 – 0.94)	0.95 (0.92 – 0.98)
E2		0.98 (0.97 – 0.99)	0.99 (0.97 – 1.00)	0.96 (0.94 – 0.97)	0.98 (0.96 – 0.99)	0.93 (0.91 – 0.94)	0.96 (0.94 – 0.98)	0.91 (0.89 – 0.92)	0.94 (0.92 – 0.96)
E3		0.99 (0.98 – 1.00)	1.01 (0.99 – 1.02)	0.98 (0.96 – 0.99)	0.99 (0.98 – 1.01)	0.95 (0.93 – 0.96)	0.97 (0.95 – 0.99)	0.93 (0.91 – 0.94)	0.95 (0.93 – 0.97)
E1	5	0.94 (0.92 – 0.95)	0.96 (0.94 – 0.97)	0.88 (0.87 – 0.89)	0.92 (0.91 – 0.93)	0.81 (0.79 – 0.82)	0.86 (0.86 – 0.88)	0.75 (0.73 – 0.77)	0.84 (0.82 – 0.86)
E2		0.94 (0.93 – 0.95)	0.96 (0.95 – 0.97)	0.89 (0.88 – 0.90)	0.93 (0.92 – 0.95)	0.82 (0.81 – 0.83)	0.89 (0.88 – 0.91)	0.73 (0.72 – 0.75)	0.83 (0.81 – 0.85)
E3		0.97 (0.96 – 0.97)	0.99 (0.98 – 1.00)	0.92 (0.91 – 0.93)	0.97 (0.95 – 0.95)	0.86 (0.85 – 0.87)	0.92 (0.91 – 0.93)	0.78 (0.77 – 0.79)	0.83 (0.82 – 0.85)
E1	10	0.91 (0.90 – 0.92)	0.94 (0.93 – 0.95)	0.83 (0.82 – 0.84)	0.89 (0.87 – 0.90)	0.72 (0.72 – 0.74)	0.81 (0.80 – 0.83)	0.63 (0.61 – 0.64)	0.74 (0.72 – 0.76)
E2		0.92 (0.92 – 0.93)	0.95 (0.94 – 0.96)	0.84 (0.84 – 0.85)	0.91 (0.90 – 0.92)	0.73 (0.73 – 0.75)	0.83 (0.83 – 0.85)	0.61 (0.60 – 0.62)	0.74 (0.73 – 0.75)
E3		0.95 (0.94 – 0.95)	0.98 (0.97 – 0.99)	0.89 (0.88 – 0.89)	0.94 (0.93 – 0.95)	0.79 (0.79 – 0.80)	0.87 (0.86 – 0.88)	0.68 (0.67 – 0.69)	0.75 (0.74 – 0.76)

[a]Logistic regression models were used to examine each of the binary trimester specific exposure effect (E1-E3) on a binary outcome (D). Model A included each trimester exposure effect separately while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U-S1} = OR_{U-S2} = OR_{U-S3} = 0.20$, and the prevalence of U = 40%

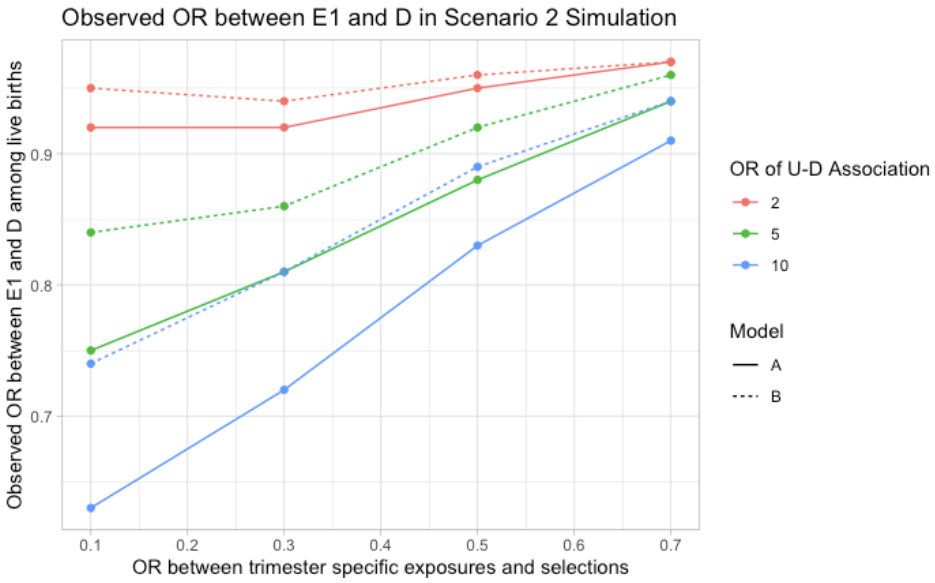

Figure 4. Observed OR between first trimester prenatal exposure (E1) and child outcome (D) in simulation of scenario 2 that assumed exposures to be marginally dependent. Model A included each trimester exposure effect separately while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U\text{-}S1} = OR_{U\text{-}S2} = OR_{U\text{-}S3} = 0.20$, $OR_{E1\text{-}E2} = OR_{E2\text{-}E3} = 8.00$ and the prevalence of U = 40%

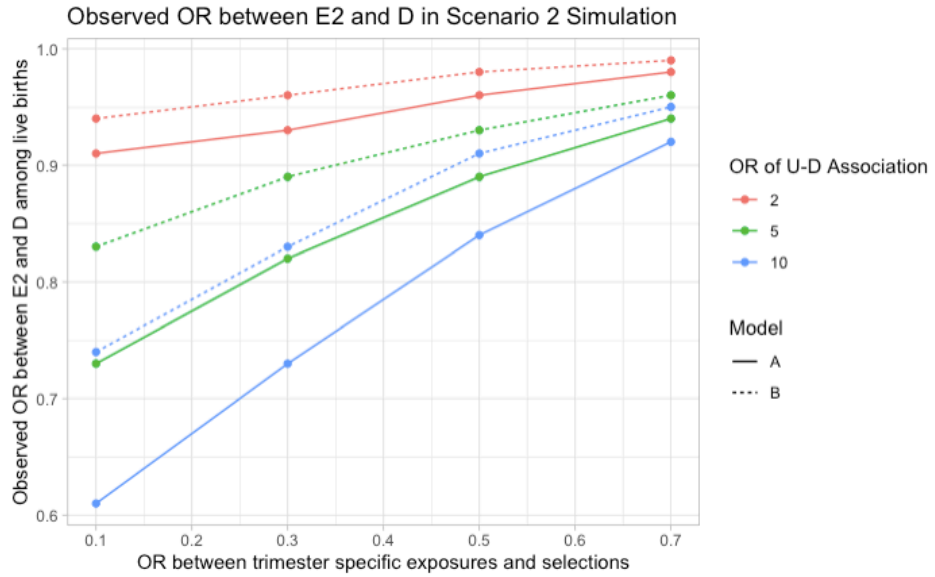

Figure 5. Observed OR between second trimester prenatal exposure (E2) and child outcome (D) in simulation of scenario 2 that assumed exposures to be marginally dependent. Model A included each trimester exposure effect separately while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U\text{-}S1} = OR_{U\text{-}S2} = OR_{U\text{-}S3} = 0.20$, $OR_{E1\text{-}E2} = OR_{E2\text{-}E3} = 8.00$ and the prevalence of U = 40%.

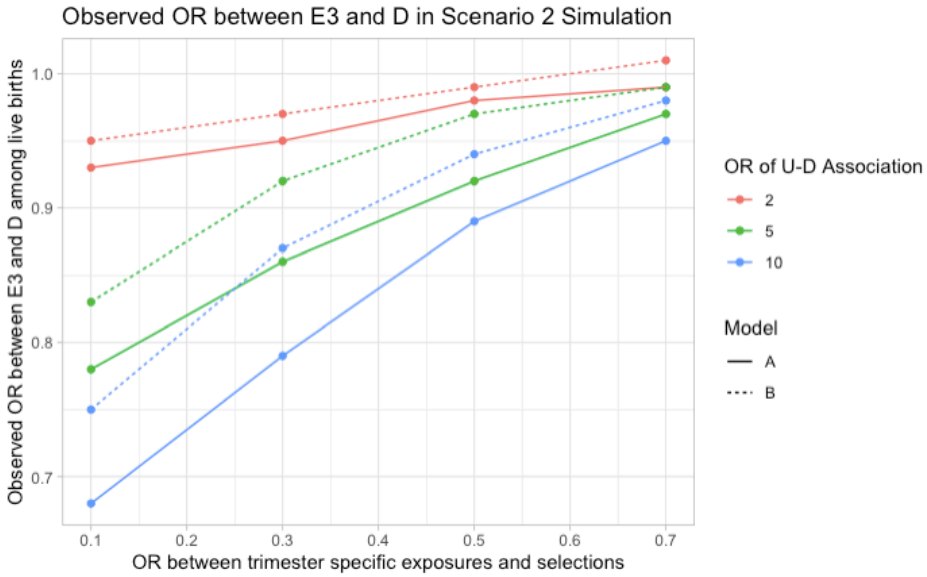

Figure 6. Observed OR between third trimester prenatal exposure (E3) and child outcome (D) in simulation of scenario 2 that assumed exposures to be marginally dependent. Model A included each trimester exposure effect separately while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U\text{-}S1} = OR_{U\text{-}S2} = OR_{U\text{-}S3} = 0.20$, $OR_{E1\text{-}E2} = OR_{E2\text{-}E3} = 8.00$ and the prevalence of U = 40%.

The estimates moved notably closer to the null (OR=1.00) when the U-D association ($OR_{U\text{-}D}$) and the exposure-selection associations ($OR_{E1\text{-}S1}$, $OR_{E2\text{-}S2}$, $OR_{E3\text{-}S3}$) decreased in strength (Figures 4 – 6). When we mutually adjusted for all three exposure variables simultaneously in model B in scenario 2, the bias sizes decreased substantially, and the effect estimates moved closer towards the null (Figures 4 – 6). However, the estimates from both models were almost identical in scenario 1 and the odds ratios curves overlapped (Figures S2 – S4, Supplemental).

In sensitivity analyses that assumed the effect of exposures on selections to be weakened from early to late gestations, the bias size was the strongest in the first-trimester pregnancy exposure variable and the weakest in the third-trimester pregnancy exposure variable (Table 6). When the association between E3 and S3 was null ($OR_{E3\text{-}S3}$), the estimate of E3 effect on D was no longer biased in model B.

Table 5. Effect estimates of trimester specific exposures in scenario 2 at varying strength[a], conditioning on live-birth cohort (S3=1) status when none of the exposures cause the outcome (true OR=1.0)[b]

	$OR_{U\text{-}D}$	$OR_{E1\text{-}S1}$ = 0.15, $OR_{E2\text{-}S2}$ = 0.50, $OR_{E3\text{-}S3}$ = 0.80		$OR_{E1\text{-}S1}$ = 0.25, $OR_{E2\text{-}S2}$ = 0.70, $OR_{E3\text{-}S3}$ = 1.00	
		Each trimester effect	Mutually adjusted trimester effect	Each trimester effect	Mutually adjusted trimester effect
E1	2	0.90 (0.89 – 0.92)	0.91 (0.90 – 0.93)	0.92 (0.91 – 0.94)	0.93 (0.92 – 0.95)
E2		0.95 (0.93 – 0.96)	0.97 (0.96 – 0.99)	0.96 (0.95 – 0.98)	0.98 (0.97 – 1.00)
E3		0.98 (0.97 – 1.00)	1.01 (0.99 – 1.02)	0.99 (0.98 – 1.00)	1.01 (1.00 – 1.02)
E1	5	0.78 (0.77 – 0.79)	0.81 (0.79 – 0.82)	0.83 (0.81 – 0.84)	0.84 (0.83 – 0.85)
E2		0.88 (0.87 – 0.89)	0.93 (0.92 – 0.95)	0.92 (0.91 – 0.93)	0.96 (0.95 – 0.98)
E3		0.94 (0.93 – 0.95)	0.99 (0.98 – 1.00)	0.97 (0.96 – 0.98)	1.01 (1.00 – 1.02)
E1	10	0.69 (0.68 – 0.70)	0.73 (0.71 – 0.74)	0.76 (0.75 – 0.77)	0.78 (0.77 – 0.79)
E2		0.83 (0.83 – 0.84)	0.91 (0.89 – 0.92)	0.89 (0.88 – 0.89)	0.95 (0.94 – 0.96)
E3		0.92 (0.91 – 0.93)	0.99 (0.98 – 1.00)	0.96 (0.95 – 0.96)	1.01 (1.00 – 1.02)

[a] The simulations assume strongest effect of exposure on selection at first trimester and weakest at third trimester.

[b] Results are rounded to 2 decimal places. Logistic regression models were used to examine individual effects of trimester specific exposures. When adjusted mutually, all three effects are included in a model. All models assumed $OR_{E1\text{-}E2} = OR_{E2\text{-}E3} = 8.0$, $OR_{U\text{-}S1} = OR_{U\text{-}S2} = OR_{U\text{-}S3} = 0.20$, $OR_{E1\text{-}S1} = OR_{E2\text{-}S2} = OR_{E3\text{-}S3} = 0.10$, and prevalence of U = 40%

DISCUSSION

In perinatal epidemiological research aiming to estimate a prenatal exposure effect on child health, studies inevitably restrict their samples to include only live born children if these health outcomes (e.g. asthma, autism spectrum disorders, etc.) are unmeasurable or unidentifiable in utero, thus resulting in biased estimates of exposure effect on outcome risk. Restricting the sample to only live born children is a form of conditioning the analytical data to a specific subset of subjects in the surviving cohort. Concerns were recently raised that this common analytical practice of excluding non-surviving fetuses in analyses when studying the effects of pregnancy exposures on long term child outcome. This form of bias has been term "live birth bias" and illustrated in a few examples. Several simulation studies found a protective relationship between smoking and preeclampsia when smoking was assumed to cause pregnancy loss, assuming a true null association between smoking and preeclampsia. The bias size strengthened when the effect of smoking on early pregnancy loss was stronger.[7,30,31] Another simulation study found a downward bias in the association between antidepressant use during pregnancy and preeclampsia when analyses conditioned on live born children, and the strength of the relationship between antidepressant use and stillbirth had a substantial impact on the bias.[6] However, previous simulations did not evaluate how live birth bias might impact the estimation of exposure effect on offspring health outcome at different times during pregnancy. Our study provided the first simulation of time-specific exposure effect on child disease risk during pregnancy when the study sample is restricted to only live born children. We also illustrated how the bias sizes changed in scenarios where exposures were assumed to be marginally independent and dependent, and how they changed based on how strongly prenatal exposures affected fetal survival at three trimesters.

Various types of selection bias in epidemiological research can be summarized using a structural approach in which they can be seen as a form of collider bias in DAGs.[20] Regardless whether the crude association between exposure and outcome is null, conditioning on the common effect of the exposure and outcome of interest (a collider) will induce a biased association between them. In general, the biased effect occurs when one of the strata of the collider is conditioned on, and that collider variable is caused directly either by the exposure or outcome themselves or directly by the cause of the exposure and the cause of the outcome. Similarly, live birth bias arises when analyses condition on the live birth status stratum of fetal survival (fetal survival = 1), a collider caused by the exposure itself as well as other unmeasured common risk factors, termed U, that are also known to cause the outcome.

This study's findings confirmed the downward bias shown in previous studies that estimated the effect of harmful exposures during pregnancy on child outcomes among live born children. Specifically, our results confirmed previous findings showing that how bias sizes increased when the effect of exposure on fetal survival at a particular trimester was stronger and when the effect of unknown or unmeasured risk factors (U) on fetal survival and child disease outcome became stronger. The prevalence of U was also found to have considerable impact on the bias size. When accounting for time-specific exposures effects, the bias sizes in model A and B were almost identical when exposures were marginally independent (scenario 1). However, when these exposures were marginally dependent (scenario 2), we observed larger bias sizes in model A compared to model B. Such difference is due to the fact that scenario 2 has more open biasing paths as a result of the dependence between the exposure variables. Mutual adjustment of all three trimester specific exposures (model B) in scenario 2, therefore, reduced the bias induced through the paths via E1 and E2, e.g. S1←E1→E2 and S2←E2→E3.

Regardless of whether the exposure variables were marginally independent (scenario 1) or dependent (scenario 2), there was a slight decreasing trend in bias size from exposure occurs in early gestation to later, assuming the strength for the exposure effect on the selection at each time was the same. One explanation for this trend could be that the simulated the proportions of fetal loss were higher in early period and gradually decreased in late gestation, reflecting the reality of pregnancy loss. Our sensitivity analyses of scenario 2 further demonstrated that as the effects of exposure on fetal survival decreased in later gestation, the bias size became weaker over time.

The sensitivity analyses of scenario 2 also showed that one of the key biased paths between each trimester exposure and the outcome is one that goes through the fetal selection variable at that trimester (S1=1, S2=1, S3=1). When third trimester exposure had no effect on live birth status ($OR_{E3\text{-}S3}$ = 1.00), all three trimester exposure estimates were biased in model A, but only the third trimester exposure (E3) estimate was not biased in model B (Table 5). When analyses conditioned on S3=1, the collider paths through S1=1 and S2=1 were also opened, thus inducing the bias at E1 and E2. Since exposures were marginally dependent, E3 was biased due to the paths through E1 and E2 in model A. When these paths were blocked due to mutual adjustment in model B, the remaining biased path from E3 was one that went through S3=1. Since E3 had no effect on S3 (e.g. $OR_{E3\text{-}S3}$ = 1.00), model B removed all bias at E3, assuming no presence of any other risk factors of E3.

Another key component of this bias structure is the group of common risk factors of fetal survival and outcome (U). Previous studies have suggested that when U were known or measured, adjusting for them would decrease the magnitude of bias.[1] For diseases or outcomes with multifactorial etiologies, the amount of unmeasured or unknown risk factors U can become extensive such that measuring and adjusting for these factors is infeasible to completely remove

live birth bias in studies of prenatal exposure effects on child outcome, conditioning on live births. Other analytic methods used to address this bias include inverse probability weighting and bias analysis using relative odds ratio. Details of these methods and their limitations have been described elsewhere.[32]

This study has a number of limitations. The scenarios assumed all subjects were recruited at conception and the timing selection we considered was limited in terms of pregnancy loss or fetal survival. In reality, women might be recruited into a pregnancy cohort study at multiple time-points with varying gestational ages of entry and pregnancy cohort studies are vulnerable to participant dropout due to reasons other than pregnancy loss. Our simulations only accounted for fetal loss during first trimester but did not consider the influence of pre-conceptional exposure on fertility. Moreover, we only tested a limited set of scenarios and limited ranges of parameters, using only binary variables and no interactions. We did not account for other types of errors, such as confounding and measurement errors. Our scenarios assumed each of the timing specific exposures had a direct effect on fetal survival at the time, but it is possible for each of them to directly affect fetal survival at later gestations. Lastly, we only considered time invariant unmeasured common causes (U) of fetal death and the outcome, excluding those that may be time-varying such as maternal BMI or blood pressure during pregnancy. Future simulation studies may consider scenarios that include other confounding variables affecting both exposures and the outcome, or common causes of the exposures and fetal survival. Continuous exposure variables that may be more reflective of the level of exposures to certain environmental exposures in the human body can also be explored in future simulations.

Our simulation findings have implications for perinatal epidemiologic studies aiming to assess timing-specific exposure effects on child disease risk. Many common prenatal exposures

have been found to persist during pregnancy such as smoking and PFAS exposure. Findings from the DNBC shows that of the women who reported ever smoking during pregnancy, only 9.5% quitted smoking during the first trimester and 2.3% quitted at the beginning of the second trimester.[27] Another analysis of the DNBC data also shows a high degree of correlation for PFAS levels between first and second trimesters (r = 0.87 for PFOS and 0.88 for PFOA). [28] Our simulation study shows that mutually adjusting for such time-specific exposure measures in one model could potentially help to mitigate live birth bias. However, multicollinearity would be a serious concern for such co-adjustment for highly correlated exposure data especially when the study sample size is small. Our study also highlights the need for improving our understanding of analytic methods to adjust for live birth bias. Since the size and magnitude of bias may vary in scenarios accounting for time-specific exposures and time-specific selections, bias adjustment methods need to consider these differences. Lastly, quantitative bias analysis for other type of effect measures, e.g. mean or risk difference for continuous or binary outcome should also be explored in future perinatal epidemiologic studies.

REFERENCES

1. Liew Z, Olsen J, Cui X, Ritz B, Arah OA. Bias from conditioning on live birth in pregnancy cohorts: an illustration based on neurodevelopment in children after prenatal exposure to organic pollutants. *Int J Epidemiol*. 2015;44(1):345-354. doi:10.1093/ije/dyu249

2. Philipp T, Kalousek DK. Generalized abnormal embryonic development in missed abortion: embryoscopic and cytogenetic findings. *Am J Med Genet*. 2002;111(1):43-47. doi:10.1002/ajmg.10476

3. Giacomucci E, Bulletti C, Polli V, Prefetto RA, Flamigni C. Immunologically mediated abortion (IMA). *J Steroid Biochem Mol Biol*. 1994;49(2-3):107-121. doi:10.1016/0960-0760(94)90001-9

4. Weselak M, Arbuckle TE, Walker MC, Krewski D. The influence of the environment and other exogenous agents on spontaneous abortion risk. *J Toxicol Environ Health B Crit Rev*. 2008;11(3-4):221-241. doi:10.1080/10937400701873530

5. Joseph KS, Kramer MS. The fetuses-at-risk approach: survival analysis from a fetal perspective. *Acta Obstet Gynecol Scand*. 2018;97(4):454-465. doi:10.1111/aogs.13194

6. Suarez EA, Landi SN, Conover MM, Jonsson Funk M. Bias from restricting to live births when estimating effects of prescription drug use on pregnancy complications: A simulation. *Pharmacoepidemiol Drug Saf*. 2018;27(3):307-314. doi:10.1002/pds.4387

7. Luque-Fernandez MA, Zoega H, Valdimarsdottir U, Williams MA. Deconstructing the smoking-preeclampsia paradox through a counterfactual framework. *Eur J Epidemiol*. 2016;31(6):613-623. doi:10.1007/s10654-016-0139-5

8. Messerlian C, Basso O. Cohort studies in the context of obstetric and gynecologic research: a methodologic overview. *Acta Obstet Gynecol Scand*. 2018;97(4):371-379. doi:10.1111/aogs.13272

9. Wilcox AJ, Weinberg CR, O'Connor JF, et al. Incidence of early loss of pregnancy. *N Engl J Med*. 1988;319(4):189-194. doi:10.1056/NEJM198807283190401

10. Bonde JP, Hjollund NH, Jensen TK, et al. A follow-up study of environmental and biologic determinants of fertility among 430 Danish first-pregnancy planners: design and methods. *Reprod Toxicol*. 1998;12(1):19-27. doi:10.1016/s0890-6238(97)00096-8

11. Wang X, Chen C, Wang L, Chen D, Guang W, French J. Conception, early pregnancy loss, and time to clinical pregnancy: a population-based prospective study. *Fertil Steril*. 2003;79(3):577-584. doi:10.1016/s0015-0282(02)04694-0

12. French FE, Bierman JM. Probabilities of fetal mortality. *Public Health Rep*. 1962;77(10):835-848.

13. Kline J, Stein ZA, Susser M, Warburton D. Smoking: a risk factor for spontaneous abortion. *N Engl J Med*. 1977;297(15):793-796. doi:10.1056/NEJM197710132971501

14. Pineles BL, Hsu S, Park E, Samet JM. Systematic Review and Meta-Analyses of Perinatal Death and Maternal Exposure to Tobacco Smoke During Pregnancy. *Am J Epidemiol*. 2016;184(2):87-97. doi:10.1093/aje/kwv301

15. Krieg SA, Shahine LK, Lathi RB. Environmental exposure to endocrine-disrupting chemicals and miscarriage. *Fertil Steril*. 2016;106(4):941-947. doi:10.1016/j.fertnstert.2016.06.043

16. Ammon Avalos L, Galindo C, Li D-K. A systematic review to calculate background miscarriage rates using life table analysis. *Birth Defects Res Part A Clin Mol Teratol*. 2012;94(6):417-423. doi:10.1002/bdra.23014

17. Toriello HV. Folic acid and neural tube defects. *Genetics in Medicine*. 2005;7(4):283-284. doi:10.1097/00125817-200504000-00009

18. Liew Zeyan, Luo Jiajun, Nohr Ellen A., et al. Maternal Plasma Perfluoroalkyl Substances and Miscarriage: A Nested Case–Control Study in the Danish National Birth Cohort. *Environmental Health Perspectives*. 128(4):047007. doi:10.1289/EHP6202

19. Greenland S, Pearl J, Robins JM. Causal diagrams for epidemiologic research. *Epidemiology*. 1999;10(1):37-48.

20. Hernán MA, Hernández-Díaz S, Robins JM. A structural approach to selection bias. *Epidemiology*. 2004;15(5):615-625. doi:10.1097/01.ede.0000135174.63482.43

21. Thompson CA, Zhang Z-F, Arah OA. COMPETING RISK BIAS TO EXPLAIN THE INVERSE RELATIONSHIP BETWEEN SMOKING AND MALIGNANT MELANOMA. *Eur J Epidemiol*. 2013;28(7). doi:10.1007/s10654-013-9812-0

22. Committee on Obstetric Practice, Society for Maternal-Fetal Medicine. Low-Dose Aspirin Use During Pregnancy. The American College of Obstetricians and Gynecologists. Accessed April 13, 2020. https://www.acog.org/en/Clinical/Clinical Guidance/Committee Opinion/Articles/2018/07/Low-Dose Aspirin Use During Pregnancy

23. Kondracki AJ, Hofferth SL. A gestational vulnerability window for smoking exposure and the increased risk of preterm birth: how timing and intensity of maternal smoking matter. *Reproductive Health*. 2019;16(1):43. doi:10.1186/s12978-019-0705-x

24. Oakeshott P, Hay P, Hay S, Steinke F, Rink E, Kerry S. Association between bacterial vaginosis or chlamydial infection and miscarriage before 16 weeks' gestation: prospective community based cohort study. *BMJ*. 2002;325(7376):1334. doi:10.1136/bmj.325.7376.1334

25. Virk J, Hsu P, Olsen J. Socio-demographic characteristics of women sustaining injuries during pregnancy: a study from the Danish National Birth Cohort. *BMJ Open*. 2012;2(4). doi:10.1136/bmjopen-2012-000826

26. Bräuner EV, Hickey M, Hansen ÅM, et al. In-utero Exposure to Maternal Stressful Life Events and Risk of Cryptorchidism: The Raine Study. *Front Endocrinol (Lausanne)*. 2019;10. doi:10.3389/fendo.2019.00530

27. Bjørnholt SM, Leite M, Albieri V, Kjaer SK, Jensen A. Maternal smoking during pregnancy and risk of stillbirth: results from a nationwide Danish register-based cohort study. *Acta Obstetricia et Gynecologica Scandinavica*. 2016;95(11):1305-1312. doi:10.1111/aogs.13011

28. Fei C, McLaughlin JK, Tarone RE, Olsen J. Perfluorinated chemicals and fetal growth: a study within the Danish National Birth Cohort. *Environ Health Perspect*. 2007;115(11):1677-1682. doi:10.1289/ehp.10506

29. Olsen J, Melbye M, Olsen SF, et al. The Danish National Birth Cohort--its background, structure and aim. *Scand J Public Health*. 2001;29(4):300-307. doi:10.1177/14034948010290040201

30. Lisonkova S, Joseph KS. Left truncation bias as a potential explanation for the protective effect of smoking on preeclampsia. *Epidemiology*. 2015;26(3):436-440. doi:10.1097/EDE.0000000000000268

31. Kinlaw AC, Buckley JP, Engel SM, Poole C, Brookhart MA, Keil AP. Left Truncation Bias to Explain the Protective Effect of Smoking on Preeclampsia: Potential, But How Plausible? *Epidemiology*. 2017;28(3):428-434. doi:10.1097/EDE.0000000000000632

32. Nohr EA, Liew Z. How to investigate and adjust for selection bias in cohort studies. *Acta Obstet Gynecol Scand*. 2018;97(4):407-416. doi:10.1111/aogs.13319

SUPPLEMENTAL

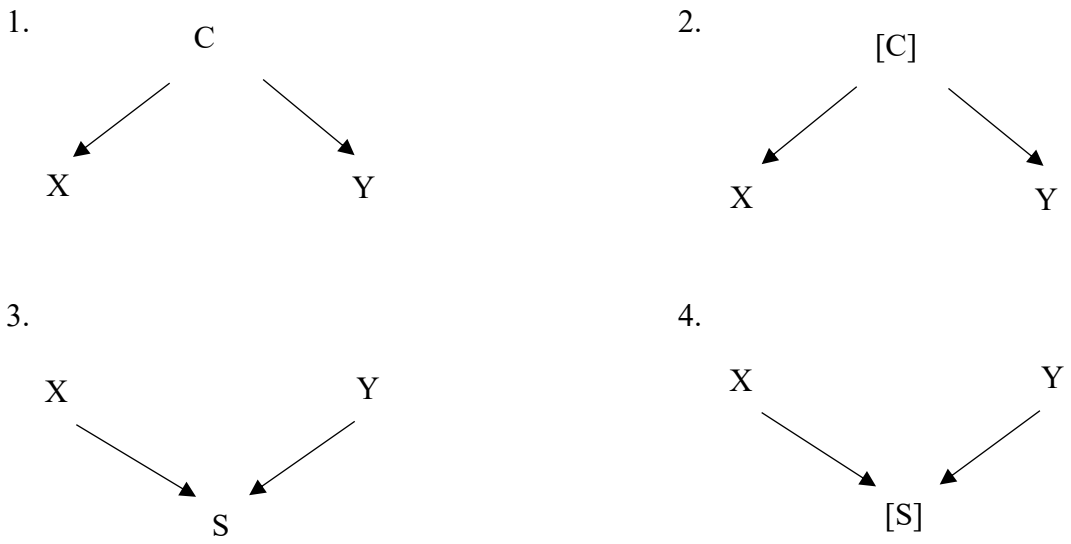

Figure S1: DAG illustrations of confounding and collider bias when X and Y are not causally related and are marginally independent. 1) There is an open backdoor confounding path through C. 2) Conditioning on C blocks the backdoor path and removes the confounding. 3) X and Y are causal determinants of S, termed a collider, that blocks the path from X to Y when left unconditioned. 4) A biasing path is opened between X and Y when S is conditioned on.

Table S1. Odds ratios and 95% simulation intervals[a] of the associations observed between the trimester specific exposures (E1-E3) on the outcome (D) in scenario 1, conditioning on live-birth cohort (S3=1) status when these exposures are marginally independent and the causal effects of all exposure variables and the outcome are null (OR=1.0), given weaker effect of U on trimester specific fetal survival ($OR_{U-S1} = OR_{U-S2} = OR_{U-S3} = 0.50$)

		$OR_{E1-S1} = OR_{E2-S2} = OR_{E3-S3} =$							
	OR_{U-D}	0.70		0.50		0.30		0.10	
		Each trimester effect	Mutually adjusted trimester effect	Each trimester effect	Mutually adjusted trimester effect	Each trimester effect	Mutually adjusted trimester effect	Each trimester effect	Mutually adjusted trimester effect
E1	2	0.99 (0.98 – 1.00)	0.99 (0.98 – 1.00)	0.97 (0.96 – 0.99)	0.97 (0.96 – 0.99)	0.97 (0.96 – 0.98)	0.97 (0.96 – 0.98)	0.95 (0.93 – 0.97)	0.95 (0.93 – 0.97)
E2		1.00 (0.99 – 1.01)	1.00 (0.99 – 1.01)	0.99 (0.98 – 1.01)	0.99 (0.98 – 1.01)	0.97 (0.96 – 0.98)	0.97 (0.96 – 0.98)	0.95 (0.93 – 0.96)	0.95 (0.93 – 0.96)
E3		1.01 (0.99 – 1.02)	1.01 (0.99 – 1.02)	1.00 (0.99 – 1.01)	1.00 (0.99 – 1.01)	0.99 (0.98 – 1.01)	0.99 (0.98 – 1.01)	0.97 (0.95 – 0.98)	0.97 (0.95 – 0.98)
E1	5	0.98 (0.97 – 0.99)	0.98 (0.97 – 0.99)	0.96 (0.95 – 0.97)	0.96 (0.95 – 0.97)	0.94 (0.92 – 0.95)	0.94 (0.92 – 0.95)	0.88 (0.86 – 0.89)	0.88 (0.86 – 0.89)
E2		0.98 (0.98 – 0.99)	0.98 (0.97 – 0.99)	0.97 (0.96 – 0.98)	0.97 (0.96 – 0.98)	0.94 (0.93 – 0.95)	0.94 (0.93 – 0.95)	0.88 (0.97 – 0.89)	0.88 (0.87 – 0.89)
E3		1.00 (0.99 – 1.01)	1.00 (0.99 – 1.01)	0.99 (0.98 – 1.00)	0.99 (0.98 – 1.00)	0.98 (0.97 – 0.99)	0.98 (0.97 – 0.99)	0.91 (0.90 – 0.92)	0.91 (0.90 – 0.92)
E1	10	0.98 (0.97 – 0.98)	0.98 (0.97 – 0.98)	0.95 (0.94 – 0.96)	0.95 (0.94 – 0.96)	0.91 (0.90 – 0.92)	0.91 (0.90 – 0.92)	0.83 (0.82 – 0.84)	0.83 (0.82 – 0.84)
E2		0.98 (0.98 – 0.99)	0.98) (0.98 – 0.99	0.96 (0.96 – 0.97)	0.96 (0.96 – 0.97)	0.92 (0.92 – 0.93)	0.92 (0.92 – 0.93)	0.84 (0.84 – 0.85)	0.84 (0.84 – 0.85)
E3		1.00 (0.99 – 1.01)	1.00 (0.99 – 1.01)	0.99 (0.98 – 0.99)	0.98 (0.98 – 0.99)	0.96 (0.95 – 0.97)	0.96 (0.95 – 0.97)	0.88 (0.87 – 0.89)	0.88 (0.87 – 0.89)

[a]Logistic regression models were used to estimate each of the binary trimester specific exposure effect (E1-E3) on a binary outcome (D). Model A included each trimester exposure effect separately, while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U-S1} = OR_{U-S2} = OR_{U-S3} = 0.20$, and the prevalence of U = 40%

Table S2. Odds ratios and 95% simulation intervals[a] of the associations observed between the trimester specific exposures (E1-E3) on the outcome (D) in scenario 1, conditioning on live-birth cohort (S3=1) status when these exposures are marginally independent and the causal effects of all exposure variables and the outcome are null (OR=1.0), with Pre(U=1) = 20%

		$OR_{E1-S1} = OR_{E2-S2} = OR_{E3-S3} =$							
	OR_{U-D}	0.70		0.50		0.30		0.10	
		Each trimester effect	Mutually adjusted trimester effect	Each trimester effect	Mutually adjusted trimester effect	Each trimester effect	Mutually adjusted trimester effect	Each trimester effect	Mutually adjusted trimester effect
E1	2	0.98 (0.96 – 0.99)	0.98 (0.96 – 0.99)	0.97 (0.96 – 0.98)	0.97 (0.96 – 0.98)	0.96 (0.94 – 0.97)	0.96 (0.94 – 0.97)	0.97 (0.95 – 0.99)	0.97 (0.95 – 0.99)
E2		0.90 (0.98 – 1.00)	0.99 (0.98 – 1.00)	0.98 (0.97 – 1.00)	0.98 (0.97 – 1.00)	0.97 (0.96 – 0.98)	0.97 (0.96 – 0.98)	0.96 (0.95 – 0.98)	0.96 (0.95 – 0.98)
E3		1.00 (0.99 – 1.01)	1.00 (0.99 – 1.01)	1.00 (0.98 – 1.01)	1.00 (0.98 – 1.01)	0.99 (0.98 – 1.00)	0.99 (0.98 – 1.00)	0.98 (0.96 – 0.99)	0.98 (0.96 – 0.99)
E1	5	0.97 (0.95 – 0.98)	0.97 (0.95 – 0.98)	0.94 (0.93 – 0.95)	0.94 (0.93 – 0.96)	0.91 (0.90 – 0.92)	0.91 (0.90 – 0.92)	0.89 (0.87 – 0.91)	0.89 (0.87 – 0.91)
E2		0.97 (0.96 – 0.98)	0.97 (0.96 – 0.98)	0.96 (0.94 – 0.97)	0.96 (0.94 – 0.97)	0.93 (0.91 – 0.94)	0.93 (0.92 – 0.94)	0.89 (0.87 – 0.90)	0.89 (0.87 – 0.91)
E3		0.99 (0.99 – 1.00)	0.99 (0.98 – 1.00)	0.98 (0.96 – 0.99)	0.98 (0.96 – 0.99)	0.96 (0.94 – 0.97)	0.96 (0.94 – 0.97)	0.90 (0.89 – 0.92)	0.90 (0.89 – 0.92)
E1	10	0.95 (0.94 – 0.96)	0.95 (0.94 – 0.96)	0.91 (0.90 – 0.92)	0.91 (0.90 – 0.92)	0.85 (0.85 – 0.87)	0.85 (0.85 – 0.87)	0.81 (0.80 – 0.83)	0.81 (0.80 – 0.83)
E2		0.96 (0.95 – 0.97)	0.96 (0.95 – 0.97)	0.92 (0.92 – 0.94)	0.93 (0.92 – 0.94)	0.88 (0.87 – 0.89)	0.88 (0.87 – 0.89)	0.81 (0.79 – 0.82)	0.81 (0.79 – 0.82)
E3		0.99 (0.98 – 1.00)	0.99 (0.98 – 1.00)	0.96 (0.95 – 0.97)	0.96 (0.95 – 0.97)	0.92 (0.91 – 0.94)	0.92 (0.91 – 0.94)	0.83 (0.82 – 0.84)	0.83 (0.82 – 0.85)

[a]Logistic regression models were used to estimate each of the binary trimester specific exposure effect (E1-E3) on a binary outcome (D). Model A included each trimester exposure effect separately, while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U-S1} = OR_{U-S2} = OR_{U-S3} = 0.20$

Table S3. Odds ratios and 95% simulation intervals[a] of the associations observed between the trimester specific exposures (E1-E3) on the outcome (D) in scenario 2, conditioning on live-birth cohort (S3=1) status when exposures are marginally dependent and the true causal effects of all exposure variables and the outcome are null (OR=1.0), given weak associations between trimester specific exposures ($OR_{E1-E2} = OR_{E2-E3} = 1.50$)

		$OR_{E1-S1} = OR_{E2-S2} = OR_{E3-S3} =$							
	OR_{U-D}	0.70		0.50		0.30		0.10	
		Each trimester effect	Mutually adjusted trimester effect	Each trimester effect	Mutually adjusted trimester effect	Each trimester effect	Mutually adjusted trimester effect	Each trimester effect	Mutually adjusted trimester effect
E1	2	0.97 (0.96 – 0.99)	0.98 (0.96 – 0.99)	0.96 (0.95 – 0.98)	0.96 (0.95 – 0.98)	0.94 (0.92 – 0.95)	0.94 (0.92 – 0.96)	0.93 (0.91 – 0.95)	0.93 (0.91 – 0.96)
E2		0.98 (0.97 – 1.00)	0.99 (0.97 – 1.00)	0.97 (0.96 – 0.99)	0.98 (0.98 – 0.99)	0.95 (0.93 – 0.97)	0.96 (0.94 – 0.97)	0.92 (0.91 – 0.94)	0.93 (0.91 – 0.95)
E3		1.01 (1.00 – 1.02)	1.01 (1.00 – 1.02)	0.99 (0.98 – 1.01)	0.99 (0.98 – 1.01)	0.97 (0.95 – 0.98)	0.97 (0.96 – 0.99)	0.94 (0.92 – 0.95)	0.94 (0.93 – 0.96)
E1	5	0.95 (0.94 – 0.96)	0.96 (0.95 – 0.97)	0.91 (0.90 – 0.92)	0.92 (0.91 – 0.93)	0.85 (0.84 – 0.87)	0.86 (0.85 – 0.87)	0.80 (0.78 – 0.82)	0.81 (0.79 – 0.83)
E2		0.96 (0.95 – 0.97)	0.96 (0.95 – 0.97)	0.92 (0.91 – 0.94)	0.93 (0.92 – 0.95)	0.87 (0.86 – 0.88)	0.88 (0.87 – 0.90)	0.79 (0.78 – 0.81)	0.81 (0.80 – 0.82)
E3		0.99 (0.98 – 1.00)	0.99 (0.98 – 1.00)	0.96 (0.95 – 0.97)	0.96 (0.95 – 0.98)	0.91 (0.90 – 0.92)	0.92 (0.90 – 0.93)	0.82 (0.81 – 0.83)	0.83 (0.82 – 0.84)
E1	10	0.93 (0.93 – 0.94)	0.94 (0.93 – 0.95)	0.89 (0.87 – 0.89)	0.89 (0.88 – 0.90)	0.79 (0.78 – 0.80)	0.80 (0.79 – 0.81)	0.70 (0.68 – 0.71)	0.71 (0.70 – 0.73)
E2		0.94 (0.94 – 0.95)	0.95 (0.94 – 0.96)	0.89 (0.88 – 0.90)	0.91 (0.90 – 0.92)	0.81 (0.80 – 0.82)	0.83 (0.82 – 0.84)	0.69 (0.68 – 0.70)	0.72 (0.70 – 0.73)
E3		0.98 (0.97 – 0.99)	0.98 (0.97 – 0.99)	0.93 (0.92 – 0.94)	0.94 (0.93 – 0.95)	0.86 (0.85 – 0.87)	0.87 (0.87 – 0.88)	0.74 (0.73 – 0.74)	0.75 (0.74 – 0.76)

[a]Logistic regression models were used to estimate each of the binary trimester specific exposure effect (E1-E3) on a binary outcome (D). Model A included each trimester exposure effect separately, while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U-S1} = OR_{U-S2} = OR_{U-S3} = 0.20$, and the prevalence of U = 40%

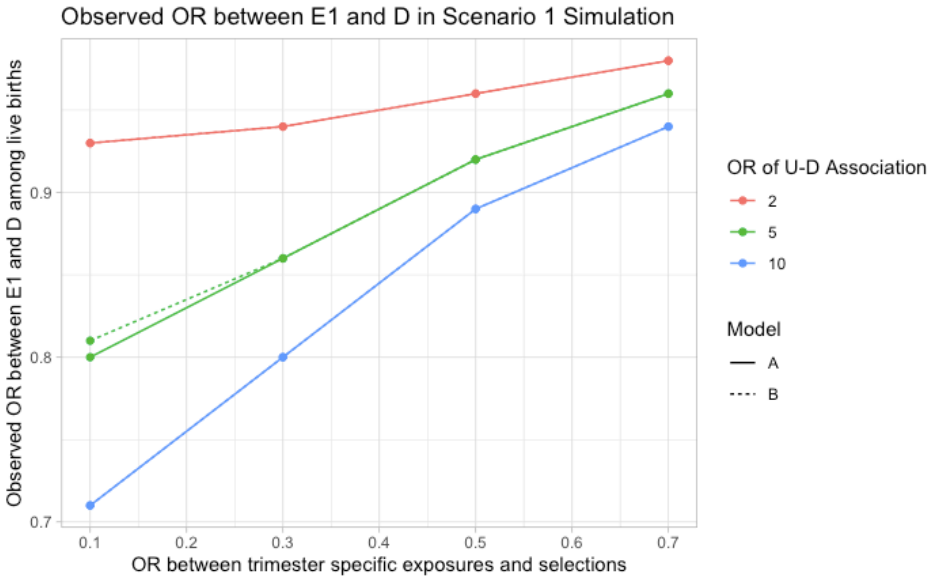

Figure S2. Observed OR between first trimester prenatal exposure (E1) and child outcome (D) in simulation of scenario 1 that assumed exposures (E1-E3) to be marginally independent. Model A included each trimester exposure effect separately while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U\text{-}S1} = OR_{U\text{-}S2} = OR_{U\text{-}S3} = 0.20$, and the prevalence of U = 40%.

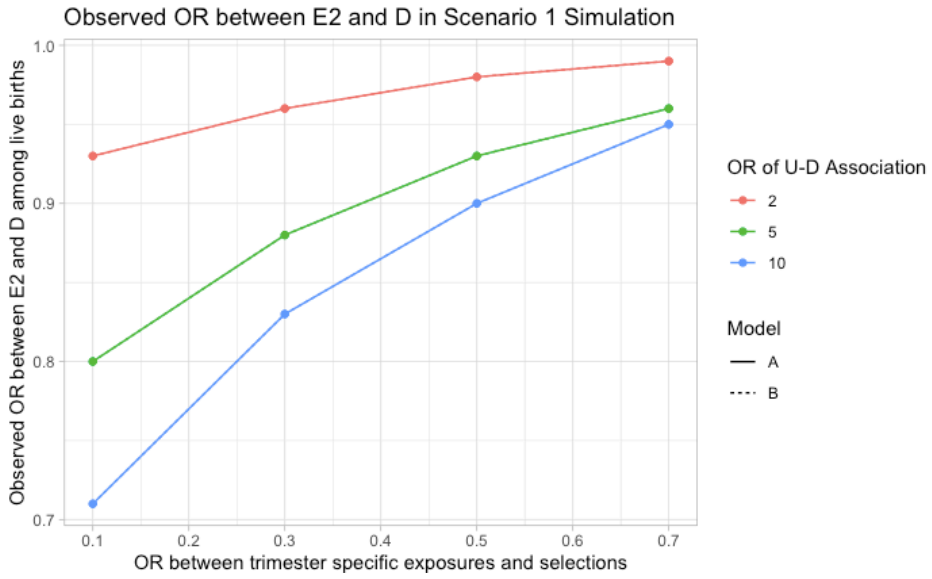

Figure S3. Observed OR between second trimester prenatal exposure (E2) and child outcome (D) in simulation of scenario 1 that assumed exposures (E1-E3) to be marginally independent. Model A included each trimester exposure effect separately while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U\text{-}S1} = OR_{U\text{-}S2} = OR_{U\text{-}S3} = 0.20$, and the prevalence of U = 40%.

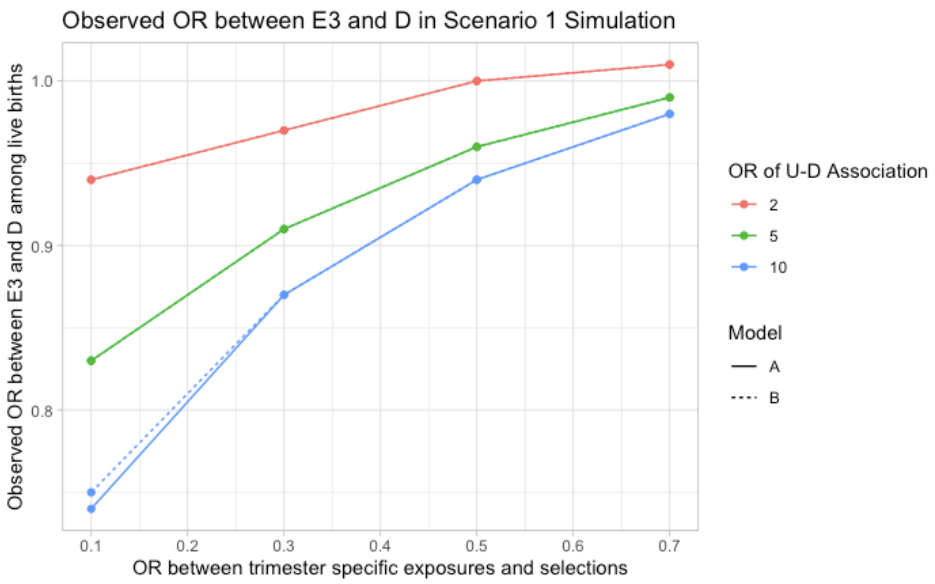

Figure S4. Observed OR between third trimester prenatal exposure (E3) and child outcome (D) in simulation of scenario 1 that assumed exposures (E1-E3) to be marginally independent. Model A included each trimester exposure effect separately while model B included all three trimester exposure variables simultaneously. All models assumed $OR_{U\text{-}S1} = OR_{U\text{-}S2} = OR_{U\text{-}S3} = 0.20$, and the prevalence of U = 40%.