Original Investigation

Maternal Smoking During Pregnancy and Offspring Conduct Problems
Evidence From 3 Independent Genetically Sensitive Research Designs

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IMPORTANCE Several studies report an association between maternal smoking during pregnancy and offspring conduct disorder. However, past research evidences difficulty in disaggregating prenatal environmental influences from genetic and postnatal environmental influences.

OBJECTIVE To examine the relationship between maternal smoking during pregnancy and offspring conduct problems among children reared by genetically related mothers and genetically unrelated mothers.

DESIGN, SETTING, AND PARTICIPANTS The following 3 studies using distinct but complementary research designs were used: The Christchurch Health and Development Study (a longitudinal cohort study that includes biological and adopted children), the Early Growth and Development Study (a longitudinal adoption-at-birth study), and the Cardiff IVF (In Vitro Fertilization) Study (an adoption-at-conception study among genetically related families and genetically unrelated families). Maternal smoking during pregnancy was measured as the mean number of cigarettes per day (0, 1-9, or ≥10) smoked during pregnancy. Possible covariates were controlled for in the analyses, including child sex, birth weight, race/ethnicity, placement age, and breastfeeding, as well as maternal education and maternal age at birth and family breakdown, parenting practices, and family socioeconomic status.

MAIN OUTCOMES AND MEASURE Offspring conduct problems (age range, 4-10 years) reported by parents or teachers using the behavior rating scales by Rutter and Conners, the Child Behavior Checklist and the Children's Behavior Questionnaire Short Form, and the Strengths and Difficulties Questionnaire.

RESULTS A significant association between maternal smoking during pregnancy and offspring conduct problems was observed among children reared by genetically related mothers and genetically unrelated mothers. Results from a meta-analysis affirmed this pattern of findings across pooled study samples.

CONCLUSIONS AND RELEVANCE Findings across 3 studies using a complement of genetically sensitive research designs suggest that smoking during pregnancy is a prenatal risk factor for offspring conduct problems when controlling for specific perinatal and postnatal confounding factors.

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Editorial

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Conduct disorder represents an issue of significant social, clinical, and practice concern, with evidence highlighting increasing rates of child conduct problems internationally.[1,2] Identifying risk factors and understanding mechanisms by which these risk factors influence conduct problems have important implications for future intervention and prevention efforts.

Maternal smoking during pregnancy is known to be a risk factor for offspring psychological problems, including attention deficits and conduct problems.[3,4] Plausible biological mechanisms have been proposed to explain the prenatal effect of nicotine exposure on neurodevelopmental processes in animals[5-7]; however, the underlying mechanisms specific to smoking in humans are not well understood.[3,8] It has been suggested that anorexigenic, hypoxic, vascular, and placental effects of nicotine may have direct teratogenic influences on the fetus and result in adverse physiological and psychological development.[9]

Longitudinal epidemiological studies have reported statistical associations between the extent of maternal smoking during pregnancy and subsequent offspring conduct disorder,[10-14] attention-deficit/hyperactivity disorder,[15,16] and criminal behavior.[17,18] Some investigations have provided evidence of a dose-response relationship between the number of cigarettes smoked during pregnancy and the rate of subsequent conduct problems in offspring.[19]

However, the effect of maternal pregnancy smoking on offspring conduct problems can be confounded by several background factors, including race/ethnicity, early age at pregnancy, low socioeconomic status, child-rearing environment, and history of maternal psychopathologic conditions.[11,13,20-23] For example, mothers who smoke during pregnancy are more likely to provide a child-rearing environment that promotes or at least condones externalizing behavior.[21] Therefore, the postnatal environment (independent of pregnancy smoking) may influence the development of conduct problems. Investigations have found that the association between maternal smoking during pregnancy and offspring conduct problems persists after accounting for these possible confounders, while others have failed to demonstrate the association when confounders were considered.[21,24]

Another problem with correlational family-based studies is the possibility of genetic risk factors and unmeasured environmental factors confounding the relationship between maternal smoking during pregnancy and offspring conduct problems.[25] Both maternal smoking during pregnancy[26] and conduct problems[27,28] are influenced by genetic factors that have been shown to overlap.[29] Maternal smoking during pregnancy is associated with externalizing problems and forming partnerships with antisocial males.[21,30,31] Moreover, adults with a history of externalizing behavior tend to provide postnatal environments that foster the transmission of this behavior across generations.[32] Indeed, passive genotype-environment correlation may be a factor in this association whereby genetic factors common to both the rearing environment (eg, harsh parenting) and the specific phenotype considered (eg, child conduct problems) underlie any observed association.[33] Thus, maternal smoking during pregnancy could be a marker of a genetic liability rather than a direct cause of children's later conduct problems. Therefore, the association between maternal smoking during pregnancy and offspring conduct problems may be genetically rather than environmentally mediated.

Recent studies using genetically sensitive designs have attempted to overcome this limitation of prior studies. Findings of studies[34-37] using sibling designs suggest that environmental variables influencing both pregnancy smoking and offspring conduct problems account for the observed associations. Previous results using an in vitro fertilization (IVF) study design, in which children are either genetically related or genetically unrelated to the mother undergoing the pregnancy,[38] and a children of twins[39] study design also suggest that unmeasured confounders indexed by inherited influences contribute to the link.

Much of the existing evidence has been obtained from studying biological parents rearing their biological children, which does not allow the effects of genetics from prenatal and postnatal environmental factors to be clearly disentangled. Nor does it allow for the role of passive genotype-environment correlation to be disentangled from genetic and postnatal environmental (eg, parenting behavior) associations.

The present study focuses on examining the links between prenatal smoking and offspring conduct problems and the contribution of psychosocial and inherited factors using data from the following 3 independent studies: the Christchurch Health and Development Study (CHDS) in New Zealand, the Early Growth and Development Study (EGDS) in the United States, and the Cardiff IVF (C-IVF) Study in the United Kingdom. In these 3 studies, data about pregnancy smoking and the behavioral outcomes have been gathered from the following: (1) in the CHDS, 1088 children reared by genetically related mothers and 36 children reared by genetically unrelated adoptive mothers; (2) in the EGDS, 310 children reared by genetically unrelated adoptive mothers; and (3) in the C-IVF Study, 636 children reared by genetically related mothers and 206 children reared by genetically unrelated mothers.

This complement of genetically sensitive research designs offers several advantages that allow advances in this important research question relative to past studies (**Table 1**). First, it allows examination of associations between maternal smoking and conduct problems in children who are reared by genetically related or genetically unrelated mothers. Second, all the studies provide information on multiple covariates, including child sex, birth weight, race/ethnicity, placement age, and breastfeeding, as well as maternal education and maternal age at birth and family breakdown, parenting practices, and family socioeconomic status. Third, results obtained from individual studies can be pooled using a meta-analytic approach to allow examination of the magnitude of common effects generated across studies. Fourth, 2 of the studies allow examination of the contribution of prenatal and possible postnatal passive genotype-environment correlation influences on the derived associations.

Methods

Sample

Study 1: CHDS

The CHDS is a longitudinal study of a birth cohort of 1265 children born in the Christchurch, New Zealand, urban region in

Table 1. Summary of Genetically Sensitive Designs of 3 Longitudinal Studies

Studies	Features	Advantage	Disadvantage
Children Reared by Genetically Related Mothers			
CHDS, C-IVF Study	Mothers provide genetic, prenatal, and postnatal environmental factors to children	Can control for several postnatal environmental factors	Cannot disentangle the effects of genetic from prenatal and postnatal environmental factors on children
Adoption-at-Birth Children Reared by Genetically Unrelated Mothers			
EGDS, CHDS	Adoptive mothers provide postnatal environmental factors but not genetic or prenatal environmental factors to children	Can test whether the effect of prenatal factors is confounded by postnatal environmental factors	Cannot remove passive genotype-environment correlation with prenatal environment influences on children
Adoption-at-Conception Children Reared by Genetically Unrelated Mothers			
C-IVF Study	Adoptive mothers provide prenatal and postnatal environmental factors but not genetic factors to children	Can test whether the effect of prenatal factors is confounded by genetic factors	Cannot disentangle the effects of prenatal and postnatal environmental influences on children

Abbreviations: CHDS, Christchurch Health and Development Study (study 1); C-IVF, Cardiff In Vitro Fertilization (study 3); EGDS, Early Growth and Development Study (study 2).

1977. Of this cohort 1124 (88.9%) were assessed on maternal smoking during pregnancy and child behavior to age 7 years. This group comprised 1088 children reared by biological mothers and 36 children reared by nonrelative adoptive mothers. The median child age at placement for adoption was 3 weeks (age range, 2-12 weeks). A detailed description of the study[40] is available elsewhere.

Study 2: EGDS

The EGDS is an ongoing, longitudinal, multisite study of linked sets of adopted children, adoptive parents, and birth parents.[41] This study drew its sample from adoption agencies from the following 4 regions in the United States: the Northwest, Southwest, Midwest, and Mid-Atlantic. The EGDS has 2 cohorts, but only data from cohort 1 were used in this study because cohort 2 does not have data at these ages yet. Cohort 1 included children who were born between 2003 and 2006 (n = 361) and were placed in nonrelative adoptive homes within 90 days of birth (median age at placement, 2 days). Birth parent data were used to assess maternal smoking, and adoptive family data were considered to evaluate the child-rearing environment (n = 311). A detailed description of the study[41] is available elsewhere.

Study 3: C-IVF Study

Children conceived via assisted reproductive technologies[42] may be genetically related to both parents (homologous IVF), the mother only (sperm donation), the father only (egg donation), or neither parent (embryo donation). Families who had a live birth between 1994 and 2002 following successful artificial reproductive treatment from any of 4 conception groups were recruited from 18 clinics in the United Kingdom and 1 US clinic.[43,44] The study design required that all donors were unrelated to either rearing parent. The numbers of families in each conception group in the full sample are 444 homologous IVF, 210 IVF with sperm donation, 175 IVF with egg donation, and 36 IVF with embryo donation. Results of comparisons among the present sample, United Kingdom national norms, and an age-matched twin sample suggest minimal differences in the mean levels of behavior.[45] Furthermore, no appreciable differences were noted among the IVF subgroups for mother-rated or father-rated adjustment problems. For the purpose of the present study, we focused on comparing mothers and children who were genetically related (homologous IVF and sperm donation) (n = 636) and those who were genetically unrelated (egg and embryo donation) (n = 206) who provided information on smoking status during pregnancy and child behavior outcomes.

Measures

Offspring Conduct Problems

In study 1 (CHDS), mothers and teachers reported on children's conduct problems at ages 6 and 7 years using selected items from the behavior rating scales by Rutter and Conners.[46] Standardized mother- and teacher-derived scores were summed for each year and then averaged over the 2 assessments to derive an overall measure of childhood conduct problems. The internal consistency of the measure was α = .76.

In study 2 (EGDS), adoptive mothers and fathers reported on children's conduct problems at ages 4½ and 6 years using the externalizing subscale of the Child Behavior Checklist[47] and the impulsivity subscale of the Children's Behavior Questionnaire Short Form.[48] Similar to the CHDS, the 2 scales were standardized and averaged at each age and then were averaged over the 2 assessments to derive an overall measure of childhood conduct problems. The internal consistency of the measure was α = .69.

In study 3 (C-IVF Study), mothers and fathers reported on children's conduct problems at ages 4 to 10 years (mean [SD] age, 5.50 [0.37] years) using the Strengths and Difficulties Questionnaire.[49] Internal consistency was acceptable (α = .67 for mothers and α = .66 for fathers).

In each study, the behavior reports have been scaled to a mean (SD) of 100 (10) within each cohort. This is to facilitate comparisons across studies.

Maternal Smoking During Pregnancy

Pregnancy smoking was reported retrospectively by mothers in all 3 studies, within 1 to 3 days of giving birth in the CHDS, at 4 months' postpartum using a life history calendar method to facilitate recall in the EGDS, and using maternal retrospective recall and antenatal records in the C-IVF Study, with reports provided by mothers during the initial assessment (chil-

dren aged ≥4 years). In study 1 (CHDS) and study 2 (EGDS), birth mothers reported on the mean number of cigarettes smoked per day in each trimester of pregnancy. In study 3 (C-IVF Study), mothers answered questions about whether they smoked 0, 1 to 9, or 10 or more cigarettes per day during pregnancy. Because the number of cigarettes smoked per day across the trimesters was highly correlated (r = 0.89 to r = 0.95 in the EGDS and r = 0.86 to r = 0.94 in the CHDS) and to make the measures comparable across the 3 studies, the maternal reports on smoking during pregnancy in studies 1 and 2 were first averaged across the trimesters and then classified into 3 levels (0, 1-9, or ≥10 cigarettes per day), thereby matching the measurement of smoking in study 3.

Covariates

To control for perinatal factors and specific characteristics of the postnatal child-rearing environment, several covariates were included in the models. These were child sex, birth weight, race/ethnicity, placement age, and breastfeeding, as well as maternal education and maternal age at birth and family breakdown, parenting practices, and family socioeconomic status (eTable in the Supplement).

Parenting Practices

In study 1 (CHDS), the maternal emotional responsiveness and avoidance of restriction and punishment subscales of the Home Observation for Measurement of the Environment Inventory[50] assessed at ages 3 to 5 years were used to measure parenting practices. The reliability of each of these scales was α = .68. In study 2 (EGDS) and study 3 (C-IVF Study), the hostility subscale of the Iowa Family Interaction Rating Scales[51] assessed parents' negative behaviors expressed toward their child. In study 2, the 5-item hostility subscale was completed by adoptive mothers and fathers when children were 27 months old and 4½ years old, and a mean score was used across both parents at both time points. In study 3, the 4-item hostility subscale was administered when children were 4 to 10 years old. Sample items include "Shout or yell at him/her because you were mad at him/her," "Criticize him/her or his/her ideas," and "Hit, push, shove, or grab him/her." Internal consistency estimates were acceptable for study 2 (α = .74) and study 3 (α = .81).

Data Analysis

The following steps were used to test whether the association between maternal smoking during pregnancy and child conduct problems was evident and still present after considering all theoretical covariates in both genetically related and genetically unrelated mother-child dyads. First, we compared the mean scores of conduct problems in children with mothers who did not smoke during pregnancy, who smoked 1 to 9 cigarettes per day, or who smoked 10 or more cigarettes per day (step 1). Second, we used ordinary least squares regression analysis to test for a significant dose-response association between maternal smoking during pregnancy and child conduct problems in each of the 3 studies. We first fitted a model containing only the maternal smoking variable as a predictor (step 2) and then assessed the potential confounding effects of child covariates, including sex, birth weight, race/ethnicity, placement age, and breastfeeding (step 3), as well as the confounding effect of maternal characteristics and postnatal environment (maternal education and maternal age at birth and family breakdown, parenting practices, and family socioeconomic status) (step 4). Steps 1 and 2 test the extent to which the associations between maternal smoking during pregnancy and child conduct problems are related for cohorts of children reared by genetically related and genetically unrelated mothers. Steps 3 and 4 control for potentially important confounders that may underlie associations across studies.

Finally, to increase the statistical power of our analyses, the regression coefficients for the genetically related samples and the genetically unrelated samples (adoption at birth) were pooled across studies using standard meta-analytic methods and assuming a random-effects model for the calculation of the pooled SE.[52] A statistical *metan* command (STATA, version 11.0; StataCorp LP) was applied to estimate the between-studies component of variance in the pooled regression analyses.

Results

Maternal Smoking During Pregnancy in the 3 Studies

The prevalence of maternal smoking during pregnancy varied across the 3 studies. In the CHDS, the prevalences of pregnancy smoking were 50.0% among children who were reared by genetically unrelated mothers and 32.7% among children who were reared by genetically related mothers. This prevalence was similar to that among the EGDS sample, with 40.8% of children having a biological mother who smoked during pregnancy. The lowest prevalences of pregnancy smoking were observed among the C-IVF Study (5.7% of children who were reared by genetically related mothers and 3.9% of children who were reared by genetically unrelated mothers).

Offspring Conduct Problems and Maternal Smoking During Pregnancy

Table 2 gives the mean scores of conduct problems in the groups of children with different rates of maternal smoking during pregnancy (0, 1-9, or ≥10 cigarettes per day) across the 3 studies. The mean scores of conduct problems were significantly different across the rates of maternal smoking among children reared by genetically related mothers (P < .001 in the CHDS and P = .005 in the C-IVF) and among children reared by genetically unrelated mothers (adoption at birth) (P = .007 in the EGDS and P = .04 in the CHDS) but not among children reared by genetically unrelated mothers (adoption at conception) (P = .98 in the C-IVF Study).

Across all the studies, for children reared by genetically related mothers and children reared by genetically unrelated mothers (adoption at birth), higher mean scores of conduct problems were observed for those whose mother smoked during pregnancy compared with those whose mother did not smoke during pregnancy. Furthermore, children whose mothers smoked 10 or more cigarettes per day had the high-

est mean scores of conduct problems. Because the sample size of the genetically unrelated mothers who smoked during pregnancy in the C-IVF Study was small (n = 8), we did not include this subgroup in further analyses because of limitations pertinent to statistical power. The correlation between the amount of smoking during pregnancy and child conduct problems varied between the C-IVF Study genetically related sample (r = 0.11, P = .005) and the CHDS genetically unrelated sample (r = 0.34, P = .04).

Association Between Maternal Smoking During Pregnancy and Child Conduct Problems

Table 3 summarizes results derived from the analysis of maternal smoking during pregnancy and child conduct problems using linear regression models (models 1-3). The unadjusted model (model 1), with maternal smoking during pregnancy as a predictor and child conduct problems score as an outcome, showed a significant association between pregnancy smoking and child conduct problems in the genetically related mother-child pairs (P < .001 in the CHDS and P = .005 in the C-IVF Study), as well as in the genetically unrelated rearing mother-child pairs (adoption at birth) (P = .007 in the EGDS and P = .04 in the CHDS).

Results of the analysis using an unadjusted model with the maximum sample size were similar to those in the samples with complete information on covariates (data are available from the corresponding author on request). The comparisons between the maximum samples and those with the full information on all covariates showed that they were not different for the frequency of pregnancy smoking or the means of child conduct problems.

In the model adjusted for child sex, birth weight, and race/ethnicity (model 2), the associations remained similar to those in the unadjusted model. The final model was adjusted for all child covariates and maternal characteristics and postnatal environment (placement age and breastfeeding, maternal education and maternal age at birth, family breakdown, parenting practices, and family socioeconomic status) (model 3). In this fully adjusted model, the association between maternal smoking during pregnancy and child conduct problems was attenuated but remained statistically significant in the genetically related mother-child pairs (P = .03 in the CHDS and P = .04 in the C-IVF Study). In the genetically unrelated rearing mother-child pairs, the association remained statistically significant in the EGDS (P = .01) but was attenuated in the CHDS (P = .12).

Results of the meta-analysis using the effect estimate (SE) from each study are also given in Table 3. These results provide further evidence for a statistical dose-specific relationship between maternal smoking during pregnancy and offspring conduct problems in both the genetically related mother-child pairs (β = 2.66, SE = 0.35, P < .001 for the unadjusted model and β = 1.13, SE = 0.56, P = .04 for the fully adjusted model) and the genetically unrelated rearing mother-child pairs (β = 2.48,

Table 2. Combined Reports of Offspring Conduct Problem Scores by Rates of Pregnancy Smoking Among Children Reared by Genetically Related Mothers and Genetically Unrelated Mothers

Study	Cigarettes per Day Smoked During Pregnancy, Mean			*r* Coefficient	*P* Value
	0	1-9	≥10		
Children Reared by Genetically Related Mothers					
CHDS	98.63 (n = 730)	100.80 (n = 160)	103.96 (n = 196)	0.21	<.001
C-IVF Study	99.20 (n = 600)	103.59 (n = 20)	104.54 (n = 16)	0.11	.005
Adoption-at-Birth Children Reared by Genetically Unrelated Mothers					
EGDS	98.66 (n = 184)	101.79 (n = 82)	102.23 (n = 45)	0.15	.007
CHDS	97.07 (n = 18)	107.81 (n = 7)	105.45 (n = 11)	0.34	.04
Adoption-at-Conception Children Reared by Genetically Unrelated Mothers					
C-IVF Study	101.43 (n = 198)	99.04 (n = 5)	103.59 (n = 3)	0.00	.98

Abbreviations: CHDS, Christchurch Health and Development Study (study 1); C-IVF, Cardiff In Vitro Fertilization (study 3); EGDS, Early Growth and Development Study (study 2).

Table 3. Combined Reports of Estimated Effects of Pregnancy Smoking on Offspring Conduct Problems for Genetically Related Rearing Mothers and Adoption-at-Birth Genetically Unrelated Rearing Mothers Before and After Adjustment for Covariates

Study	Model 1, Unadjusted		Model 2, Adjusted for Child Covariates		Model 3, Fully Adjusted	
	β Level (95% CI)	*P* Value	β Level (95% CI)	*P* Value	β Level (95% CI)	*P* Value
Children Reared by Genetically Related Mothers						
CHDS	2.61 (1.88 to 3.33)	<.001	2.36 (1.59 to 3.08)	<.001	0.82 (0.08 to 1.56)	.03
C-IVF Study	3.07 (0.95 to 5.18)	.005	3.00 (0.86 to 5.15)	.006	2.15 (0.11 to 4.18)	.04
Pooled	2.66 (1.97 to 3.34)	<.001	2.58 (1.33 to 3.82)	<.001	1.13 (0.02 to 2.24)	.04
Adoption-at-Birth Children Reared by Genetically Unrelated Mothers						
EGDS	2.08 (0.57 to 3.59)	.007	2.20 (0.57 to 3.83)	.008	1.99 (0.48 to 3.90)	.01
CHDS	4.51 (0.32 to 8.70)	.04	4.17 (−0.22 to 8.56)	.07	4.27 (−0.90 to 9.44)	.12
Pooled	2.48 (0.72 to 4.23)	.006	2.44 (0.91 to 3.96)	.002	2.17 (0.72 to 3.62)	.003

Abbreviations: CHDS, Christchurch Health and Development Study (study 1); C-IVF, Cardiff In Vitro Fertilization (study 3); EGDS, Early Growth and Development Study (study 2).

SE = 0.90, P = .006 for the unadjusted model and β = 2.17, SE = 0.74, P = .003 for the fully adjusted model).

Discussion

Results derived from the present study showed that maternal smoking during pregnancy was associated with offspring conduct problems. This association was observed for children reared by both genetically related and genetically unrelated mothers. In the genetically unrelated (adoption at birth) mother-child pairs, the characteristics of an adoptive mother and the child-rearing environment are distinct from the presence or absence of pregnancy smoking. Therefore, our results suggest that the association between maternal smoking during pregnancy and offspring conduct problems was not confounded by maternal characteristics or the child-rearing environment, specifically parenting practices. Moreover, this association was observed when a possible passive genotype-environment correlation was removed using the attributes of the adoption-at-birth design (EGDS and CHDS adoptees).

Our findings add to evidence highlighting the adverse effects of smoking during pregnancy as a risk factor for offspring conduct problems. First, results of prior sibling design studies[34-36] suggest that siblings who differed in their exposure to pregnancy smoking did not differ for conduct problems across childhood and adolescence. However, these studies were not able to control for a passive genotype-environment correlation, whereas our study included an adoption-at-birth design and could demonstrate that having a postnatal environment free from genetic confounding did not explain the association between maternal smoking during pregnancy and offspring conduct problems. Second, prenatal exposure to smoking might represent an inherited rather than a true environmental risk factor underlying offspring conduct problems.[38,39] It is possible that preexisting genetically based differences in the propensity to engage in externalizing behavior may confound the relationship between maternal smoking during pregnancy and offspring conduct problems.[53] For example, a previous study by Rice et al[38] using data from the C-IVF Study showed that the association between prenatal smoking and child antisocial behavior was observed in genetically related but not in genetically unrelated mother-child pairs, suggesting that the association represents an inherited rather than a truly causal effect.

Results from previous studies using the IVF design[38] and a children-of-twins design[39,54] suggest that a passive genotype-environment correlation may contribute to the link between maternal smoking during pregnancy and offspring conduct problems. Combined with existing research, findings from the present study demonstrate that the underlying mechanisms for the association between maternal pregnancy smoking and offspring conduct problems are present during the prenatal period. These may involve common genetic factors that may interact with pregnancy smoking. Results of recent molecular genetic studies[55-58] revealed that offspring with a particular genetic profile are more sensitive to the negative effect of maternal smoking during pregnancy than those without. For example, a gene × environment interaction between the *COMT* and *MAOA* genes and maternal smoking during pregnancy on offspring aggressive behavior has been reported.[56,57] Most important, the interaction between *COMT* and pregnancy smoking might be explained at the epigenetic level because the association of nicotine exposure with methylation of the gene promoter has recently been demonstrated.[59] To further our knowledge of the effects of maternal smoking during pregnancy on offspring conduct problems, genetically sensitive designs incorporating information on genetic and epigenetic markers are needed in future studies.

Our study has strengths and limitations. Findings provided in the present study were obtained by using comparable measures of maternal smoking during pregnancy across the 3 studies. There is a possibility that our results are affected by historical smoking trends, specifically in relation to the CHDS. However, any bias due to cohort effects is likely to be minimal because results are consistent across studies. Multiinformant reports (from a mother and a father or from a mother and a teacher) were used to measure child conduct problems. These measures are not identical, yet the pattern of findings is consistent across independent samples of mother-child pairs derived from distinct geographical and social backgrounds when controlling for a wide range of possible covariates. In addition, we confirmed the substantive findings in the pooled data sets using meta-analysis. Given that each design has its own set of strengths and weaknesses, different designs were used. Indeed, as Rutter[60] outlines, greater confidence is achieved when there is convergence of findings across studies using a complement of research designs.

Strengths notwithstanding, several limitations of the study should be noted. First, the number of smokers in the genetically unrelated group in study 3 (C-IVF Study) was small (n = 8), thereby precluding incorporation of this group in the regression analysis and meta-analysis. Second, the prevalence of maternal smoking during pregnancy among the C-IVF Study genetically related sample was significantly lower than that among the CHDS (5.7% vs 32.7%). However, the magnitude of association between maternal smoking during pregnancy and conduct problems was similar in these 2 distinct sample groups before adjustment for potential confounders (β = 2.61 in the CHDS and β = 3.07 in the C-IVF). Third, exposure to other substances (drugs and alcohol) during pregnancy, as well as postnatal smoking exposure (passive smoking) following birth, may be important risk factors for child development and need to be considered in future studies. As an additional test, we examined the role of passive smoking or environmental tobacco smoke if this measure was available (CHDS). Results remained unchanged when we incorporated this measure into the analysis (data are available from the corresponding author on request). Fourth, our study (like most in the field) predominantly relied on maternal self-report of smoking. Although such methods have been shown to have excellent agreement with antenatal records,[35,46] biological measures may provide more accurate quantitative data concerning the true levels of nicotine that the fetus was exposed to during pregnancy. Also, future studies may need to explore a time-specific effect of exposure to pregnancy smoking.

In conclusion, using a complement of genetically sensitive research designs, the present study examined the relationship between maternal smoking during pregnancy and offspring conduct problems among children reared by genetically related and genetically unrelated mothers when controlling for specific perinatal and postnatal factors. Our findings suggest an association between pregnancy smoking and child conduct problems that is unlikely to be fully explained by postnatal environmental factors (ie, parenting practices) even when the postnatal passive genotype-environment correlation has been removed. The causal explanation for the association between smoking in pregnancy and offspring conduct problems is not known but may include genetic factors and other prenatal environmental hazards, including smoking itself. Research designs that allow disaggregation of genetic and environmental pathways underlying intergenerational transmission of psychopathologic conditions are critical for understanding the role of maternal smoking during pregnancy and could have important implications for future intervention and prevention programs aimed at remediating child conduct problems.

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