Published by Oxford University Press on behalf of the International Epidemiological Association
International Journal of Epidemiology 2010;39:1531–1540
doi:10.1093/ije/dyq133

Prenatal smoking exposure and offspring stress coping in late adolescence: no causal link

Ralf Kuja-Halkola,[1,2]* Brian M D'Onofrio,[3] Anastasia N Iliadou,[1] Niklas Långström[1,2] and Paul Lichtenstein[1]

[1]Department of Medical Epidemiology and Biostatistics, Karolinska Institutet, Stockholm, Sweden, [2]Centre for Violence Prevention, Karolinska Institutet, Stockholm, Sweden and [3]Department of Psychological and Brain Sciences, Indiana University, Bloomington, IN, USA

*Corresponding author. Department of Medical Epidemiology and Biostatistics, Karolinska Institutet, PO Box 281, SE-171 77 Stockholm, Sweden. E-mail: ralf.kuja-halkola@ki.se

Accepted 1 July 2010

Background *In utero* exposure to tobacco smoking has been suggested to cause persistent alterations in cognitive functioning. We examined if mothers' smoking during pregnancy (SDP) is associated with long-term impairment in offspring stress coping and the causal mechanism behind a possible link.

Methods We used a large cohort ($n = 187\,106$) of young males in Sweden (mean age = 18.2 years), who underwent a semi-structured psychological assessment in 1997–2006, including an evaluation of stress coping ability, as part of the compulsory military conscript examination. We compared differentially exposed siblings within nuclear families and cousins in extended families and used multilevel structural equation models to disentangle genetic from environmental contributions to the association between SDP and stress coping.

Results SDP and offspring stress coping was moderately strongly associated when comparing unrelated individuals [regression coefficient (b) = −0.38 on a nine-point scale; 95% confidence interval (CI) −0.40 to −0.36, $P < 0.0001$]. In contrast, it disappeared when siblings were compared ($b = 0.11$; 95% CI −0.01 to 0.23, $P = 0.071$). This familial confounding was entirely due to genetic influences.

Conclusions SDP is an established risk factor for pregnancy- and birth-related complications. However, we found no long-term effect of SDP on offspring stress coping. Rather, the observed association was due to familial confounding of genetic origin; women prone to SDP also transmit genes to their children that are associated with poorer coping with stress.

Keywords Smoking during pregnancy, adolescent stress coping, children-of-sibling model, intergenerational association

Introduction

Maternal smoking during pregnancy (SDP) has been linked to several negative perinatal outcomes, such as low offspring birth weight,[1–5] preterm birth,[1,4–6] spontaneous abortion[4,5,7] and sudden infant death syndrome.[4,5,7,8] In addition, long-lasting behaviour problems have been suggested; for example, compared with unrelated controls, offspring who experienced SDP have increased risk of externalizing

problems[9] including attention-deficit/hyperactivity disorder,[7,9] aggression,[10] criminality,[7,11] poorer general cognitive functioning[12] and poorer academic and intellectual performance.[13–15]

Birth-related SDP outcomes (e.g. lower birth weight, preterm birth) appear causal,[5] whereas emerging evidence suggests that the link to most studied long-term behaviours is confounded by other risks or unmeasured familial effects, such as shared environmental or genetic risks.[9,11–16]

One aspect of individual development that recently received substantial interest is the ability to cope with stress. For instance, increased stress vulnerability has been observed as a consequence of prenatal nicotine exposure in rats.[17] Animal studies also suggest that prenatal nicotine exposure increases locomotor activity and causes learning and memory problems.[7] Specifically, the fetal programming hypothesis[18,19] includes suggestions that the major regulatory systems involved in stress responses, the autonomic nervous system and the hypothalamic–pituitary–adrenal axis, could be permanently altered early in life; both pre- and post-natally.[18,19] These alterations could, for example, be caused by restricted access to food or specific nutrients, maternal adversity or exposure to synthetic glucocorticoids (e.g. cortisol).[19] Birth weight has often been used as a proxy for measuring adverse fetal environment,[18,19] and prior Swedish studies suggested that offspring size at birth is related to the measure of stress susceptibility used in the current study.[20–22]

Other possible teratogenic SDP-related mechanisms include disturbed placenta function and impaired transport of nutrients and oxygen to the fetus,[7] and nicotine-caused intrauterine hypoxia or birth asphyxia[7] leading to fetal death or long-term neurological deficits, or cellular alterations to the central and peripheral nervous systems.[7,23] The decrease in 'fetal breathing' (fetal thoracic movements), reported to occur after nicotine exposure, causes lung maturation to slow down and reduces the blood flow to the fetus.[7] Additionally, activation of neurotransmitter receptors in the fetal brain could lead to epigenetic alterations involving permanent change in cell functioning that might not be detected until much later in the more developed, adolescent, brain.[7,24]

However, another important mechanism through which SDP could effect offspring behaviour is the passing of genetic vulnerability from parent to offspring; a passive gene–environment correlation.[25] In effect, repeated results from studies of behavioural problems (e.g. externalizing behaviour,[9] criminality[11] and poor academic achievement[14,15]) in offspring exposed to SDP were later found to be entirely confounded by familial risks. That is, a selective mechanism for SDP exposure exists so that mothers who smoke during pregnancy share also other risk factors with their children; hence, these other risk factors cause the observed adverse outcomes rather than SDP *per se*. Variables such as maternal age, education or socio-economic status might be the source of this selection,[7] but also unmeasured familial environment similarity and genetic risks.

We aimed to investigate whether the association between intrauterine exposure to SDP and stress coping in Swedish late adolescent men persisted after controlling for measured and unmeasured confounding caused by intrafamilial similarity.

Methods

Study population

We linked several nationwide longitudinal registries, maintained by government agencies in Sweden, using the unique personal identification number given to all Swedish citizens. We used data from the Multi-Generation[26] and Education Registers,[27] the 1990 Swedish Census,[28] and the Conscript,[29] Medical Birth,[30,31] Total Population and National Crime Registers.[32] Eligible for the study were all male youth in Sweden who underwent an evaluation regarding suitability for duty by a clinical psychologist at compulsory conscription for military services during 1997–2006 and born 1982–88 (SDP registration at antenatal care started in 1982). For this study, we used data collected regarding stress coping ($n = 187\,106$). Military conscription was mandatory for Swedish men until 2008 and enforced by law. The majority of conscripts were 18 years old [79.3%; mean age = 18.2 years, standard deviation (SD) = 0.4, median = 18.2, range 17.1–24.3 years]. Individuals were linked to their siblings and cousins via parents and grandparents using the Multi-Generation Register, thus identifying extended and nuclear families. This register links all children born 1932 or later in Sweden to both their parents. Nuclear families were indexed by the mothers (164 563 mothers with at least one child), whereas extended families were indexed by the maternal or paternal grandmother. There were 150 268 extended families with at least one individual in the offspring generation. Offspring with both maternal and paternal cousins could be included in two extended families.

Exposure

Starting in 1982, all pregnant women in contact with public tax-funded antenatal care in Sweden are asked by their personal midwife about SDP; this information is included in the Medical Birth Register. The coverage is excellent; >98% of all births are recorded in the register.[30] SDP data were available for 162 371 of the 187 106 (86.8%) pregnancies in the study. Of these, 44 550 women reported SDP (27.4%), which is comparable with earlier findings.[5,9,11,12] The validity of self-reported SDP is high in general[33] and previous studies suggest good validity also in the current sample.[2,34]

Outcome

A clinical psychologist rated individual psychological functioning (PF) at conscription, purportedly reflecting stress coping during wartime,[20,35] based on a standardized, 20–25-min semi-structured interview. PF was rated 1–9 on a nine-point Likert-type scale; higher values indicate better coping. The distribution was stipulated to be normal with mean = 5 and SD = 2 (χ^2 goodness-of-fit test with nine categories; $P = 0.23$, indicating no reason to reject the normality assumption). The individual PF score was used as a proxy for general ability to cope with stress.

Covariates

We adjusted our analyses using offspring, nuclear family and extended family confounders and mediators.

Offspring confounders

Maternal age was divided into five categories: <20, 20–24, 25–29, 30–34 and >34 years. Birth year was used alone and together with Conscript Register data to stratify age at conscription into categories; ⩽17.50, 17.51–18.50 and >18.50 years. A birth order variable for male nuclear family offspring was also constructed.

Offspring mediators

We considered two mediators, both obtained from the Medical Birth Register: gestational time divided into categories: <32, 32–36, 37–41 and >41 weeks; and birth weight.

Parental confounders

Parental occupation, divided into seven categories,[36] income and cohabitation status were all based on the 1990 Census. The Register of Education for 2004 provided highest parental educational level, classified into seven categories.[27] Parental criminal convictions for 1973–2004 were collected from the National Crime Register. The Total Population Register supplied mother's country of birth divided into 12 categories according to geographic and demographic similarities. Finally, we included a variable indicating if a half-sibship existed for offspring within the nuclear family.

Extended family confounders

We also included a variable on whether an individual had maternal or paternal half-cousins.

Statistical methods

To analyse the effect of SDP on PF, we used linear regression treating PF as a normally distributed variable. Results are presented both crude (unadjusted) and adjusted for possible confounders. To handle possible period effects, we adjusted the crude unrelated and cousin models for birth year and the crude sibling model for birth order.

Since birth weight and gestational age could be mediators of the association between SDP and PF, additional analyses were run to investigated whether these covariates mediated the association between SDP and PF as proposed by the fetal programming hypothesis.

Since we aimed at isolating a possible direct effect of SDP on PF by eliminating possible familial confounding, we ran additional models to test if familial effects distorted the association. With these, we compared PF in siblings and cousins differentially exposed to SDP to explore if the association remained when looking at within-family effects (i.e. if differentially exposed siblings/cousins also differed in PF). Hence, we used the extended family and nuclear family as clusters and sub-clusters to capture similarities within families. These analyses were performed with hierarchical linear models (HLM)[37,38] using SAS Proc Mixed.[39] Thus, unmeasured variables common to individuals in the nuclear or extended family (i.e. shared genes and environments) were accounted for.[9] We call this approach the Children-of-Siblings model since it is similar to the statistical methods used when examining variables for children of twins in conjunction with variables for their parents, or the Children-of-Twins model.[9,37,40,41] SDP and continuous covariates were centred around the cluster means (for both nuclear and extended family) which yielded covariates equivalent to fixed effects.[15] Furthermore, this procedure reduced possible bias due to correlation between covariates and residual errors.[42] We utilized an informed backwards elimination process when deciding which covariates to use; thus, these may differ across models.

We performed sibling–sibling and cousin–cousin comparisons on two different data subsets. Siblings were compared using a subset consisting of two siblings within a nuclear family. Nuclear families were solely indexed by mothers, since 91% of children in Sweden stays with their mother when parents divorce or separate,[43] and our explicit aim was to capture possible familial effects. There were 26 118 individual siblings within 13 059 nuclear families. Cousin comparisons were made on a subset consisting of two cousins within an extended family. There were 52 888 individuals from 47 684 nuclear families within 26 421 extended families included in the subset (individuals eligible for comparisons within two extended families might be included twice in the analysis). To examine the difference between how SDP influence PF in full- and half-sibling/cousins, we conducted the analyses separately. The concordance of SDP in sibling and cousin pairs are presented in Table 1.

Finally, we aimed to disentangle the source of the familial confounding of the association between maternal SDP and her son's PF by partitioning the

Table 1 Concordance regarding exposure to maternal smoking during pregnancy in sibling and cousin pairs among all male children born 1982–88 in Sweden and assessed for PF at age 18 years as part of mandatory military conscript evaluation

Relation	SDP = 0	SDP = 1	Total
Full siblings			25 452
Concordant	18 702	4450	23 152
Discordant	1150	1150	2300
Half siblings			666
Concordant	256	292	548
Discordant	59	59	118
Full cousins			50 038
Concordant	27 480	5106	32 586
Discordant	8726	8726	17 452
Half cousins			2850
Concordant	978	492	1470
Discordant	690	690	1380

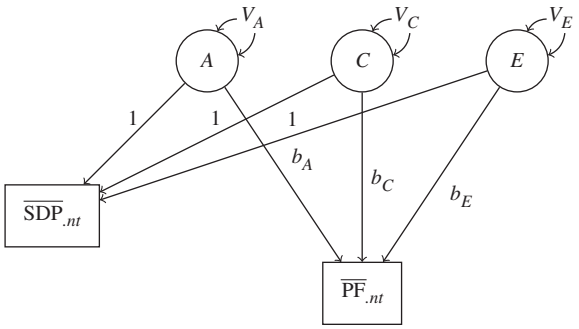

Figure 1 Variance partitioning for *A*, genetics; *C*, the shared environment and *E*, non-shared environment. The graph represents the model for one of two sibling mothers. SDP: mean smoking during pregnancy exposure; PF: mean psychological functioning capacity (stress coping); *A*: latent variable representing the genes; *C*: latent variable representing the shared environment; *E*: latent variable representing the non-shared environment; V_A: variance of latent variable *A*; V_C: variance of latent variable *C*; V_E: variance of latent variable *E*; b_A: regression coefficient for PF regressed on *A*; b_C: regression coefficient for PF regressed on *C*; b_E: regression coefficient for PF regressed on *E*

variance of the intergenerational association in a two-level hierarchical structural equation model (SEM) using the statistical software program Mplus.[44,45] Analyses were first performed with PF, and then on the residuals of PF retrieved from a linear regression model using the covariates. Because the results of these two analyses were very similar, we present only the analyses of the former. Results from the latter analyses are presented in the Supplementary Figure S1 (Supplementary data are available at *IJE* online). Wherever possible, we picked one pair of sisters and their children for each extended family (24 468 children from 11 485 sister pairs). The two SEM levels refer to within-mothers, comparing the association between mothers' SDP and offspring's PF within each of the nuclear families, and between-mothers, comparing the average SDP association with the average SDP between nuclear families. The variance was partitioned into three parts *A*, *C* and *E*, corresponding to genetics, shared environment (makes siblings similar) and non-shared environment (makes siblings different)[9] (Figure 1). The partitioning of the variance comes from taking into account the genetic relatedness between the mothers; full siblings share 50% of their co-segregating genes, while half-siblings share 25%. This notion was incorporated in the SEM as constraints on the genetic variance parameter modelled (V_A in Figure 1). Another constraint was that the modelled shared environment variance parameter V_C was equal within full sibling pairs and maternal half-siblings, whereas this parameter was set to 0 for paternal half-siblings (again, because 91% of the children remain with their mother when parents separate).[43] This way of modelling allows us to draw conclusions about which of the intergenerational paths (b_A, b_C and b_E) that explain the association between SDP and PF. The method, as applied in Children-of-Twins model, has been described elsewhere.[9,41]

Results

Mean PF scores are presented in Table 2 for SDP and offspring covariates and in Table 3 for parental covariates.

The crude estimate of the association of SDP with PF was −0.38 [95% confidence interval (CI) −0.40 to −0.36, $P < 0.0001$; Figure 2, for results from the regression analysis: Supplementary Table S1 (Supplementary data are available at *IJE* online)]. This means that a child exposed to SDP had, on average, a PF or stress coping score 0.38 points lower on the nine-point scale than an unrelated child unexposed to SDP. The following analyses explored the mechanisms behind this negative association.

Each of the included covariates individually predicted PF (analyses not shown). When adjusting for these, the association was attenuated [$b = -0.15$; 95% CI −0.18 to −0.13, $P < 0.0001$; Figure 2 and Supplementary Table S2 (Supplementary data are available at *IJE* online)].

Having adjusted for known confounders, we investigated if the association was due to unmeasured familial confounding by performing sibling and cousin analyses with HLM. The crude and adjusted between extended family estimates ('unrelated' estimates) remained approximately similar (Figure 2). Thus, offspring in extended families where SDP had occurred also had lower mean PF scores, regardless of if a mother smoked during his own or somebody else's

Table 2 Offspring characteristics for all 187 106 male children born 1982–88 in Sweden and assessed for PF at age 18 years as part of mandatory military conscript evaluation

Characteristic	*n* (%)	PF, mean (SD)
Smoking during pregnancy		
Yes	44 550 (23.8)	4.6 (1.8)
No	117 822 (63.0)	5.0 (1.8)
Missing	24 734 (13.2)	4.9 (1.8)
Gestational time, weeks		
28–31	654 (0.3)	4.5 (1.7)
32–36	8966 (4.8)	4.8 (1.8)
37–41	162 873 (87.4)	4.9 (1.8)
>41	13 812 (7.4)	4.9 (1.8)
Missing	801 (0.4)	4.7 (1.7)
Age at conscription, years		
⩽17.50	2245 (1.2)	5.0 (1.7)
17.51–18.50	158 625 (84.8)	4.9 (1.8)
>18.50	26 236 (14.0)	4.6 (1.9)
Year of birth		
1982	29 971 (16.0)	4.8 (1.8)
1983	28 231 (15.1)	4.9 (1.8)
1984	25 915 (13.9)	5.1 (1.7)
1985	30 243 (16.2)	4.8 (1.9)
1986	27 017 (14.4)	4.8 (1.8)
1987	25 801 (13.8)	4.7 (1.8)
1988	19 928 (10.7)	4.9 (1.7)
Birth order		
1	169702 (90.7)	4.9 (1.8)
2	16 812 (9.0)	4.9 (1.8)
3	578 (0.3)	4.8 (1.8)
4	10 (0.0)	4.5 (1.5)
5	1 (0.0)	7 (−)
Missing	3 (0.0)	4.3 (2.1)
Birth weight, kg		
<1.50	1641 (0.9)	4.7 (1.7)
1.50–1.99	1006 (0.5)	4.6 (1.8)
2.00–2.49	3503 (1.9)	4.6 (1.8)
2.50–2.99	16 233 (8.7)	4.7 (1.8)
3.00–3.49	55 210 (29.5)	4.8 (1.8)
3.50–3.99	68 784 (36.8)	4.9 (1.8)
⩾4	40 729 (21.8)	4.9 (1.8)
Mother's age at delivery, years		
<20	5491 (2.9)	4.3 (1.8)
20–24	43 338 (23.2)	4.7 (1.8)
25–29	69 977 (37.4)	4.9 (1.8)
30–34	47 551 (25.4)	5.0 (1.8)
>34	20 749 (11.1)	4.9 (1.8)
Total	187 106	4.9 (1.8)

pregnancy (sibling's or a cousin's). As seen in Figure 2, family-adjusted effects (from the sibling and cousin models) differed from 'unrelated' effects. When accounting for covariates for SDP-discordant siblings, the within-regression effect of SDP on PF completely disappeared ($b = 0.13$; 95% CI −0.01 to 0.27, $P = 0.068$; Figure 2), providing strong evidence for familial confounding. Another way to address the mechanism behind this would be to study also half-siblings, but because of too few half-siblings discordant for SDP (59 pairs, Table 1) the statistical power was minimal [Supplementary Table S6 (Supplementary data are available at *IJE* online)]. When full cousins were studied, the adjusted within-regression parameter was close to zero ($b = -0.05$; 95% CI −0.11 to 0.01, $P = 0.073$; Figure 2) and reduced compared with the within-half-cousins effect ($b = -0.23$; 95% CI −0.44 to −0.02, $P = 0.030$; Figure 2) and to the between extended family parameter, again indicating substantial familial confounding. For full tables of regression coefficients from sibling and cousin analyses, see Supplementary Tables S3–S10 (Supplementary data are available at *IJE* online).

We found minimal influence of birth weight and gestational age on the association between SDP and PF, suggesting that these covariates did not mediate neither the crude nor the adjusted association that we could verify. This held true for unrelated comparisons as well as sibling and cousin comparisons (analyses not shown).

Because the HLM analyses indicated substantial familial confounding, we tried to estimate genetic and environmental effects on the association using SEM. When fitting the *ACE* model, both the intergenerational paths b_C and b_E had very large standard errors [$b_A = -1.48$, standard error (SE) = 0.90, $P = 0.10$; $b_C = -1.31$, SE = 2.65 $P = 0.62$; $b_E = 0.18$, SE = 0.41, $P = 0.66$], which indicates that one latent variance parameter in the model is negligible.[41] To test whether the *ACE* model fitted the data better than the *AE* and/or the *CE* model, two scaled-difference χ^2-tests[46] were performed. We found no evidence that the *ACE* model explained the data better than the *AE* model [$\chi^2 = 1.2$, degrees of freedom (df) = 2, $P = 0.56$]. In contrast, when comparing the *ACE* model with the *CE* model, the result was in favour of the *ACE* model ($\chi^2 = 11.2$, df = 2, $P = 0.004$). We used the Bayesian Information Criterion (BIC) to determine which of the models *ACE* and *AE* that fitted the data best. The *AE* model outperformed the *ACE* ($BIC_{AE} = 126\,493$, $BIC_{ACE} = 126\,511$; The Akaike Information Criterion (AIC) yielded a similar result: $AIC_{AE} = 126\,277$, $AIC_{ACE} = 126\,280$). Therefore, in subsequent models the shared environment parameters were set to zero.[9,41] Results from the model fitting are presented in Figure 3. The regression coefficient b_A expressing the genetic intergenerational transmission on PF was

Table 3 Characteristics for parents of all 187 106 male children born 1982–88 in Sweden and assessed for PF at age 18 years as part of mandatory military conscript evaluation

Characteristic	Maternal characteristics		Paternal characteristics	
	n (%)	PF, mean (SD)	*n* (%)	PF, mean (SD)
Parent's occupation				
Unskilled blue-collar worker	48 941 (26.2)	4.6 (1.7)	34 349 (18.4)	4.6 (1.8)
Skilled blue collar	20 845 (11.1)	4.8 (1.7)	37 737 (20.2)	4.7 (1.7)
Low-level white collar	28 390 (15.2)	5.0 (1.7)	16 086 (8.6)	5.0 (1.8)
Intermediate-level white collar	36 426 (19.5)	5.2 (1.8)	32 521 (17.4)	5.1 (1.8)
High-level white collar	13 395 (7.2)	5.3 (1.8)	28 976 (15.5)	5.2 (1.8)
Self employed	5615 (3.0)	5.0 (1.8)	14 889 (8.0)	4.9 (1.8)
No information/ uncategorized	10 576 (5.7)	4.7 (1.8)	9692 (5.2)	4.7 (1.8)
Missing[a]	22 918 (12.2)	4.6 (1.8)	12 856 (6.9)	4.5 (1.8)
Parent's income, Swedish kronor				
<100 000	91 964 (49.2)	4.7 (1.8)	25 538 (13.6)	4.5 (1.8)
100 000–199 900	89 125 (47.6)	4.9 (1.8)	96 755 (51.7)	4.7 (1.8)
200 000–299 900	5194 (2.8)	5.4 (1.8)	50 536 (27.0)	5.1 (1.8)
300 000–399 900	686 (0.37)	5.4 (1.8)	10 059 (5.4)	5.4 (1.7)
⩾400 000	137 (0.07)	5.6 (2.0)	4218 (2.3)	5.6 (1.8)
Parent's highest education at childbirth				
<9 years	4179 (2.2)	4.3 (1.8)	10 649 (5.7)	4.5 (1.7)
9 years	17 955 (9.6)	4.4 (1.8)	26 099 (13.9)	4.6 (1.7)
1–2 years upper secondary education	67 436 (36.0)	4.7 (1.7)	61 131 (32.7)	4.7 (1.8)
3 years upper secondary education	23 762 (12.7)	4.9 (1.8)	23 859 (12.8)	5.0 (1.8)
<3 years post-secondary education	33 622 (18.0)	5.1 (1.8)	25 175 (13.5)	5.2 (1.8)
>3 years post-secondary education	36 056 (19.3)	5.2 (1.8)	29 013 (15.5)	5.2 (1.8)
Postgraduate education	992 (0.5)	5.3 (1.9)	2862 (1.5)	5.2 (1.8)
Missing	3104 (1.7)	4.5 (1.9)	8318 (4.4)	4.5 (1.9)
Parent convicted of a criminal offence[b]				
No	167 303 (89.4)	4.9 (1.8)	117 597 (62.9)	5.0 (1.8)
Yes	19 803 (10.6)	4.5 (1.8)	69 509 (37.1)	4.6 (1.8)
Mother's country of birth				
Sweden	170 017 (90.9)	4.9 (1.8)	n/a	n/a
Scandinavia except Sweden	7466 (4.0)	4.6 (1.8)	n/a	n/a
25 European Union member states except Scandinavia and former Eastern Europe	1367 (0.7)	4.7 (1.8)	n/a	n/a
Former Eastern Europe	1745 (0.9)	4.7 (1.9)	n/a	n/a
Europe except Scandinavia, 25 European Union member states and former Eastern Europe	2439 (1.3)	4.4 (1.7)	n/a	n/a

(continued)

Table 3 Continued

Characteristic	Maternal characteristics		Paternal characteristics	
	n (%)	PF, mean (SD)	*n* (%)	PF, mean (SD)
Former USSR	113 (0.1)	4.6 (1.8)	n/a	n/a
Africa	507 (0.3)	4.4 (1.6)	n/a	n/a
Canada or USA	261 (0.1)	4.7 (1.9)	n/a	n/a
Rest of North America	64 (0.0)	4.7 (1.8)	n/a	n/a
South America	814 (0.4)	4.4 (1.7)	n/a	n/a
Asia	2165 (1.2)	4.3 (1.7)	n/a	n/a
Oceania	35 (0.0)	4.5 (1.4)	n/a	n/a
Missing	113 (0.1)	4.7 (1.7)	n/a	n/a
Half-sibship[c]			n/a	n/a
No	186 157 (99.5)	4.9 (1.8)	n/a	n/a
Yes	949 (0.5)	4.5 (1.8)	n/a	n/a
Cohabitation status			n/a	n/a
Parents cohabiting	159 102 (85.0)	4.9 (1.8)	n/a	n/a
Parents not cohabiting	7998 (4.3)	4.5 (1.8)	n/a	n/a
Missing	20 006 (10.7)	4.9 (1.8)	n/a	n/a

[a]Missing values from the 1990 Census was due to the reason that some Swedes failed to respond as required by law.
[b]Criminal convictions were obtained from the national crime registry; hence, there were no missing data.
[c]The low prevalence of half-siblings was due to the short time interval (7 years, 1982–88), during which both the index children and the half-siblings had to be born, and that only male children with a score for PF at conscription were included.
n/a: not applicable; either only maternal values are present or there is only one value per nuclear family.

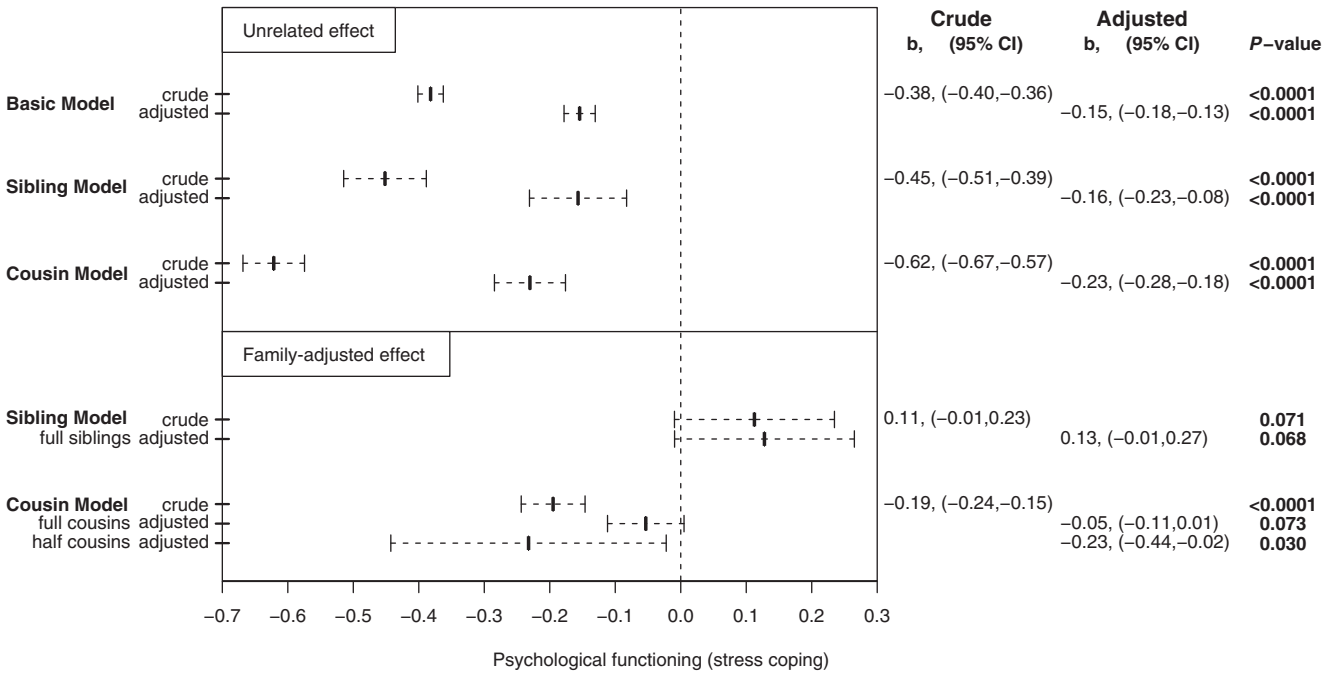

Figure 2 Regression coefficient estimates and 95% CIs for PF as a function of maternal smoking during pregnancy among male offspring born 1982–88 in Sweden and assessed for PF at age 18 years as part of mandatory military conscript evaluation

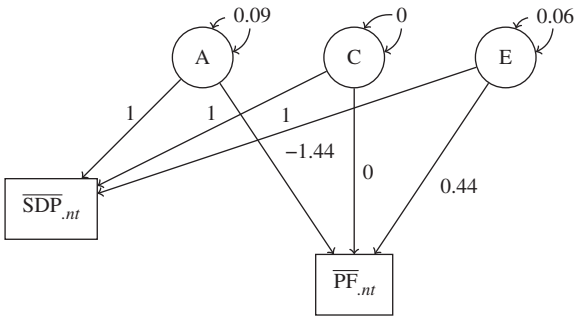

Figure 3 Variance partitioning for *A*, genetics; *C*, the shared environment and *E*, non-shared environment. The graph represents the fitted model for one of two sibling mothers. SDP: mean smoking during pregnancy exposure; PF: mean psychological functioning capacity (stress coping); *A*: latent variable representing the genes; *C*: latent variable representing the shared environment; *E*: latent variable representing the non-shared environment, variance of latent variable $A = 0.09$ ($P < 0.0001$), variance of latent variable $E = 0.06$ ($P < 0.0001$), regression coefficient for PF regressed on $A = -1.44$ ($P < 0.0001$), regression coefficient for PF regressed on $E = 0.44$ ($P = 0.037$)

negative ($b_A = -1.44$, SE = 0.13, $P < 0.0001$) while the non-shared environmental regression coefficient was positive ($b_E = 0.44$, SE = 0.21, $P = 0.037$). For the full model, see Supplementary Figure S2 (Supplementary data are available at *IJE* online).

Discussion

We aimed to investigate the effect of intrauterine exposure to SDP on offspring stress coping in late adolescence. We used nationwide longitudinal registers, to compare not only unrelated individuals differentially exposed to SDP, but also relatives (siblings and cousins) to explore possible familial confounding and estimate the roles of genetic and environmental determinants.

Our main finding was that the observed association between SDP and poorer PF was entirely confounded by familial factors. Since the association could not be entirely explained by selected a priori confounders, we applied models that investigated unmeasured confounding based on similarities within nuclear and extended families. Familial confounding was evident; the association between SDP and poorer offspring stress coping decreased when half-cousin comparisons were used instead of unrelated individuals, and disappeared completely in within full-cousin and full-sibling comparisons. A possible reason for mothers to change smoking habits between pregnancies is if a life-altering event has occurred. However, a Swedish study on whether smoking habit changes after an adverse pregnancy outcome found only modest effects on continued smoking in next pregnancy.[47] Thus, this is probably not a major reason for the familial confounding, especially since such effects are even less influential in the comparison between smoking discordant sisters. Additionally, data suggested that genetic effects entirely accounted for this familial confounding. The present results concur with previous studies in humans, suggesting that associations between SDP and cognitive/behavioural outcomes in adolescent offspring are not causal but subject to substantial familial confounding,[9,11–15] primarily due to genetic rather than environmental mechanisms. One possible mechanism is that mothers transmit smoking liability to offspring and offspring smoking influenced stress reactivity. We could not test this since no data on smoking were available for the conscripts. Regardless, to be informative, studies of the effect of SDP and other parental risk factors on offspring must take familial confounding into account.

We also tested the fetal programming hypothesis, the study of which often used low birth weight as a proxy for adverse fetal environment.[18,19] When comparing siblings or cousins differently exposed to SDP, the inclusion of birth weight as a potential mediator of the link between SDP and offspring PF, the latter remained essentially unchanged. Thus, either the lowering of birth weight due to SDP is not in the same causal pathway as the effect on PF or the effect of low birth weight on PF is also due to familial confounding.

Our study had several strengths, particularly its size and longitudinal total population-based design with high coverage of exposure data and outcome. As supported by Swedish data, we assumed that children are primarily raised by their biological mothers when parents divorce or separate.[43] Since PF was assessed by professionals employed by the Swedish armed forces, the rating was classified. Hence, although previous studies used PF as a stress coping measure,[20–22,35] we could not explicitly validate it. As indirect support, however, the test has been used for several decades, and it has been validated in that it correlates with military rank at the completion of military service.[48] Regarding exposure, we used mothers' smoking status at prenatal care registration (approximately the first trimester) as SDP measure. Self-reported SDP might be less reliable in later years, since the stigma associated with smoking while pregnant has increased. However, studies support its validity,[2,33,34] the period when SDP was measured was quite short (1982–88), and we controlled for period effects when including birth year/birth order as a covariate. We cannot differentiate between prenatal only and prenatal plus postnatal smoking; therefore, the associations examined could be due to postnatal smoking as well as SDP. Another limitation of the study is that analyses

were done in men, hence generalization to women cannot be assumed.

Finally, we want to stress that mothers' SDP is associated with numerous adverse outcomes, especially related to birth and infancy.[5,49] However, our results add to the accumulating evidence that SDP have no, or only minor, long-term causal effects on offspring cognitive functioning.[9,11–15] Other factors accounts for the association, and in the case of stress coping the confounds seem to be mainly of genetic origin. To conclude, mothers prone to SDP also transmit genes to their children, which cause poorer stress coping in the latter.

Supplementary Data

Supplementary data are available at *IJE* online.

Funding

Swedish National Prison and Probation R&D; the Swedish Research Council – Medicine; the Indiana University Faculty Research Support Program, NARSAD; the National Institute of Child Health and Human Development (grant number HD056354).

Conflict of interest: None declared.

KEY MESSAGES

- Fetal exposure to tobacco smoking is suggested to cause long-term alterations to regulatory systems involved in stress response.
- We found an association between SDP and worse stress coping in male offspring at 18 years of age.
- This association is not causal but subject to substantial familial confounding.
- The familial confounding seems to be primarily of genetic origin.

References

[1] Andres RL, Day MC. Perinatal complications associated with maternal tobacco use. *Semin Neonatol* 2000;**5:**231–41.

[2] Cnattingius S, Haglund B. Decreasing smoking prevalence during pregnancy in Sweden: the effect on small-for-gestational-age births. *Am J Public Health* 1997;**87:**410–13.

[3] Hardy JB, Mellits ED. Does maternal smoking during pregnancy have a long-term effect on the child? *Lancet* 1972;**2:**1332–36.

[4] The 2004 United States Surgeon General's Report. The health consequences of smoking. *NSW Public Health Bull* 2004;**15:**107.

[5] Cnattingius S. The epidemiology of smoking during pregnancy: smoking prevalence, maternal characteristics, and pregnancy outcomes. *Nicotine Tob Res* 2004;**6(Suppl 2):** S125–40.

[6] Kyrklund-Blomberg NB, Cnattingius S. Preterm birth and maternal smoking: risks related to gestational age and onset of delivery. *Am J Obstet Gynecol* 1998;**179:** 1051–55.

[7] Ernst M, Moolchan ET, Robinson ML. Behavioral and neural consequences of prenatal exposure to nicotine. *J Am Acad Child Adolesc Psychiatry* 2001;**40:**630–41.

[8] Haglund B, Cnattingius S. Cigarette smoking as a risk factor for sudden infant death syndrome: a population-based study. *Am J Public Health* 1990;**80:**29–32.

[9] D'Onofrio BM, Van Hulle CA, Waldman ID *et al*. Smoking during pregnancy and offspring externalizing problems: an exploration of genetic and environmental confounds. *Dev Psychopathol* 2008;**20:**139–64.

[10] Huijbregts SC, Seguin JR, Zoccolillo M, Boivin M, Tremblay RE. Associations of maternal prenatal smoking with early childhood physical aggression, hyperactivity-impulsivity, and their co-occurrence. *J Abnorm Child Psychol* 2007;**35:**203–15.

[11] D'Onofrio BM, Singh AL, Iliadou A *et al*. Familial confounding of the association between maternal smoking during pregnancy and offspring criminality: a population-based study in Sweden. *Arch Gen Psychiatry* 2010;**67:** 529–38.

[12] Kafouri S, Leonard G, Perron M *et al*. Maternal cigarette smoking during pregnancy and cognitive performance in adolescence. *Int J Epidemiol* 2009;**38:** 158–72.

[13] Lundberg F, Cnattingius S, D'Onofrio BM *et al*. Maternal smoking during pregnancy and intellectual performance in young adult Swedish male offspring. *Paediatr Perinatal Epidemiol* 2010;**24:**79–87.

[14] Lambe M, Hultman C, Torrång A, Maccabe J, Cnattingius S. Maternal smoking during pregnancy and school performance at age 15. *Epidemiology* 2006;**17:** 524–30.

[15] D'Onofrio BM, Singh AL, Iliadou A *et al*. A quasi-experimental approach of maternal smoking during pregnancy and offspring academic achievement. *Child Dev* 2010;**81:** 80–100.

[16] Knopik VS. Maternal smoking during pregnancy and child outcomes: real or spurious effect? *Dev Neuropsychol* 2009;**34:**1–36.

[17] Vaglenova J, Birru S, Pandiella NM, Breese CR. An assessment of the long-term developmental and behavioral teratogenicity of prenatal nicotine exposure. *Behav Brain Res* 2004;**150:**159–70.

[18] Phillips DI. Programming of the stress response: a fundamental mechanism underlying the long-term effects of the fetal environment? *J Intern Med* 2007;**261:**453–60.

[19] Phillips DI, Jones A. Fetal programming of autonomic and HPA function: do people who were small babies have enhanced stress responses? *J Physiol* 2006;**572:**45–50.

[20] Nilsson PM, Nyberg P, Östergren PO. Increased susceptibility to stress at a psychological assessment of stress tolerance is associated with impaired fetal growth. *Int J Epidemiol* 2001;**30:**75–80.

[21] Nilsson PM, Nilsson JA, Östergren PO, Rasmussen F. Fetal growth predicts stress susceptibility independent of parental education in 161991 adolescent Swedish male conscripts. *J Epidemiol Community Health* 2004;**58:** 571–73.

[22] Lundgren EM, Cnattingius S, Jonsson B, Tuvemo T. Intellectual and psychological performance in males born small for gestational age. *Horm Res* 2003;**59 (Suppl 1):**139–41.

[23] Slotkin TA. Fetal nicotine or cocaine exposure: which one is worse? *J Pharmacol Exp Ther* 1998;**285:**931–45.

[24] Huizink AC, Mulder EJ. Maternal smoking, drinking or cannabis use during pregnancy and neurobehavioral and cognitive functioning in human offspring. *Neurosci Biobehav Rev* 2006;**30:**24–41.

[25] Rutter M. Proceeding from observed correlation to causal inference. *Perspectives Psychol Sci* 2007;**2:**377–95.

[26] *Multi-generation Register 2005 — A Description of Contents and Quality*. Örebro: Statistics Sweden, 2006. http://www.scb.se/statistik/_publikationer/BE9999_2005A01_BR_BE96ST0606.pdf (Accessed July 19, 2010).

[27] Utbildningsregistret. Statistics Sweden. Available from: http://www.scb.se/statistik/UF/UF0506/_dokument/UF0506_DO_2004.pdf (Accessed July 19, 2010).

[28] *Folk- och bostadsräkningen 1990, FoB 90*. Stockholm: Statistics Sweden, 1999. http://www.scb.se/statistik/BE/BE0205/_dokument/BE0205_DO_1990.pdf (Accessed July 19, 2010).

[29] Magnusson PK, Gunnell D, Tynelius P, Davey Smith G, Rasmussen F. Strong inverse association between height and suicide in a large cohort of Swedish men: evidence of early life origins of suicidal behavior? *Am J Psychiatry* 2005;**162:**1373–75.

[30] *The Swedish Medical Birth Register — A Summary of Content and Quality*. Stockholm: Centre for Epidemiology, 2003. http://www.socialstyrelsen.se/publikationer2003/2003-112-3 (Accessed July 19, 2010).

[31] Cnattingius S, Ericson A, Gunnarskog J, Källén B. A quality study of a medical birth registry. *Scand J Soc Med* 1990; **18:**143–48.

[32] Fazel S, Grann M. The population impact of severe mental illness on violent crime. *Am J Psychiatry* 2006; **163:**1397–403.

[33] Lindqvist R, Lendahls L, Tollbom Ö, Åberg H, Hakansson A. Smoking during pregnancy: comparison of self-reports and cotinine levels in 496 women. *Acta Obstet Gynecol Scand* 2002;**81:**240–44.

[34] Hatziandreu EJ, Pierce JP, Fiore MC *et al*. The reliability of self-reported cigarette consumption in the United States. *Am J Public Health* 1989;**79:**1020–23.

[35] Leboeuf-Yde C, Larsen K, Ahlstrand I, Volinn E. Coping and back problems: analysis of multiple data sources on an entire cross-sectional cohort of Swedish military recruits. *BMC Musculoskelet Disord* 2006;**7:**39.

[36] *Socioekonomisk indelning (SEI) 1982*. Stockholm: Statistics Sweden. http://www.scb.se/Grupp/Hitta_statistik/Forsta_Statistik/Klassifikationer/_Dokument/SEI-MIS.pdf (Accessed July 19, 2010).

[37] Harden KP, Lynch SK, Turkheimer E *et al*. A behavior genetic investigation of adolescent motherhood and offspring mental health problems. *J Abnorm Psychol* 2007; **116:**667–83.

[38] Raudenbush SW, Bryk AS. *Hierarchical Linear Models Applications and Data Analysis Methods*. 2nd edn. Thousand Oaks, CA: Sage Publications, 2002.

[39] Littell RC, Milliken GA, Stroup WW, Wolfinger RD, Schnabenberger O. *SAS for Mixed Models*. Cary, NC: SAS Press, 2006.

[40] D'Onofrio BM, Turkheimer E, Emery RE *et al*. A genetically informed study of marital instability and its association with offspring psychopathology. *J Abnorm Psychol* 2005;**114:**570–86.

[41] D'Onofrio BM, Turkheimer EN, Eaves LJ *et al*. The role of the children of twins design in elucidating causal relations between parent characteristics and child outcomes. *J Child Psychol Psychiatry* 2003;**44:**1130–44.

[42] Neuhaus JM, McCulloch CE. Separating between- and within-cluster covariate effects by using conditional and partitioning methods. *J Roy Stat Soc* 2006;**68:**859–72.

[43] *Fakta om den svenska familjen*. *Demografiska rapporter (1994:2)*. Stockholm: Statistics Sweden, 1994.

[44] Muthén BO. *Mplus Technical Appendices*. Los Angeles, CA: Muthén & Muthén, 1998–2004.

[45] Muthén LK, Muthén BO. *Mplus User's Guide*. 5 edn. Los Angeles, CA: Muthén & Muthén, 1998–2007.

[46] Satorra A, Bentler PM. A scaled difference chi-square test statistic for moment structure analysis. *Psychometrika* 2001;**66:**507–14.

[47] Cnattingius S, Akre O, Lambe M, Ockene J, Granath F. Will an adverse pregnancy outcome influence the risk of continued smoking in the next pregnancy? *Am J Obstet Gynecol* 2006;**195:**1680–86.

[48] Carlstedt B. *Validering av inskrivningsprövningen mot vitsord från den militära grundutbildningen*. Karlstad: Försvarshögskolan, Klara AB Tryckeri i Karlstad, 1999.

[49] Johansson AL, Dickman PW, Kramer MS, Cnattingius S. Maternal smoking and infant mortality: does quitting smoking reduce the risk of infant death? *Epidemiology* 2009;**20:**590–97.