

Interactive Effects of in Utero Nutrition and Genetic Inheritance on Cognition: New Evidence Using Sibling Comparisons¹

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Abstract: A large literature links early environments and later outcomes, such as cognition; however, little is known about the mechanisms. One potential mechanism is sensitivity to early environments that is moderated or amplified by the genotype. With this mechanism in mind, a complementary literature outside economics examines the interaction between genes and environments, but typically do not use strong research designs, resulting in problems of endogeneity and bias in estimation. A key issue in the literature is exploring environmental variation that is not exogenous, which is potentially problematic if there are gene-environment correlation or gene-gene interactions. This paper explores the importance of this issue by extending previous findings of an interaction between the *FADS2* gene, which is associated with the processing of essential fatty acids related to cognitive development, and early life nutrition in explaining later-life IQ. Using sibling pairs with genetic data in the Wisconsin Longitudinal Study we extend a previous, and widely cited, gene-environment study that explores an interaction between the *FADS2* gene, which is associated with the processing of essential fatty acids related to cognitive development, and early life nutrition in explaining later-life IQ. Our base OLS findings suggest that individuals with “favorable” *FADS2* variants obtain 2.5 IQ points for each standard deviation increase in birth weight, our measure of the early nutrition environment; while, individuals without the favorable variants of *FADS2* do not have a statistically significant association with early nutrition, implying the genotype is influencing the effects of environmental exposure. When including family-level fixed effects, however, the magnitude of the gene-environment interaction is reduced by half and statistical significance dissipates, implying the significant interaction between *FADS2* and early nutrition in explaining later life IQ may be due to unobserved, family-level factors. The example has wider implications for the practice of investigating gene-environment interactions when the environmental exposure is not exogenous and robustness to unobserved variation in the genome is not controlled for in the analysis.

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1. Introduction

It is well known that intellectual development is a product of both genetic and environmental factors. In particular, early nutrition (including in utero) has lifelong effects on a range of health and economic outcomes. Evidence from the Dutch Hunger Winter (Stein 1975) has shown that famine conditions suffered in utero led to increases in adult obesity and mental illness. Related findings in the economics literature have shown that birth weight differences are associated with long term differences in IQ, education, and earnings (Black et al. 2007). Additionally, Doyle et al. (2009) and Heckman et al. (in press) argue that the early childhood period may be the optimal time for interventions associated with ameliorating SES differences. An important insight from the economics literature, not largely employed outside economics, is the need to control for shared family environments when estimating the impacts of early conditions. For example, Almond et al. (2005) show large reductions (>80%) in estimate of the effects of birth weight on mortality and medical expenditures when sibling differences are employed. Oreopoulos et al. (2008) show similar sensitivity to some estimates based on controlling for family environments, particularly for siblings in the relationship between birth weight and later-life test scores.

Understanding the mechanisms behind the links between early environments and later outcomes has been the subject of increasing research across several disciplines. In particular, potential interactions between the “nature” and “nurture” domains has been an increasingly common direction that has linked social and biological sciences and has led to novel findings that suggest focusing on “nature” or “nurture” in isolation misses important channels determining intellectual development. In other words, the genetic endowment of an individual has the potential to moderate or amplify the effects of a given environment. In terms of policy, this interplay between genes and environments gives rise to differential effects of environmental interventions, from which targeted interventions based on the genome may provide added efficiency both in terms of cost and desired outcomes. Therefore, it is necessary to more robustly examine the causal relationship between gene-environment interactions in economic and social outcomes of interest.

A key investigation along these lines of gene-environment interaction is from Caspi et al (2007), who show a replicated interaction effect between early nutrition, as measured by breastfeeding, and a specific genetic variant thought to modify dietary fatty acids, which itself is potentially important in cognitive development. In particular, the authors interact two genetic variations in the *FADS2* gene with breastfeeding measures to predict childhood IQ outcomes. They find that, in two different study populations, individuals carrying the GG genotype of SNP rs174575 had no advantage or disadvantage

from breastfeeding while those with at least one C-allele had a large (6.4 point) IQ advantage over individuals who were not breastfed.² Although the authors check for common confounding influences, the potential that the genetic variants were correlated with the environmental exposure or that there could be gene-gene interaction rather than gene-environment interaction remains.

In replication studies, however, the interaction between FADS2 and birth weight has been questioned. Steer et al. (2010) find the GG genotype of SNP rs174575 has a significant interaction with breastfeeding in determining IQ—the opposite of the allele, or gene variant, considered within Caspi et al. This inconsistency between the findings of Steer et al. and Caspi et al. also carries over to SNP rs1535. However, a statistically significant association remains between FADS2 and birth weight, implying the candidate gene-environment interaction is plausible. Going further, an additional attempt at replication is unable to produce the significant interactions found in Caspi et al. (Martin et al. 2011).

Our approach is not tied to the direct replication of Caspi et al. Rather, we consider a more general and widely used proxy for early nutrition, birth weight, in order to explore an alternative interactive channel for FADS2. As argued below, the mechanism connecting FADS2 and breastfeeding is also potentially connected to birth weight. As validity for this substitution, we find a positive and statistically significant interaction between birth weight and each variant of FADS2 considered by Caspi et al. The use of this extension, however, is to test the robustness of the interaction to the potential confounding effects of unobserved family-level omitted variables.

Our approach is broad, intending to provide a check for robustness for statistically significant interactions between candidate genes and environments. Previous studies, found outside economics that have focused on the potential interactions between a phenotype's genetic endowment and the effect of endowments in varied environments have been unable to fully control for omitted variables (i.e., unobserved genetic and environmental effects) that may lead to a spurious GxE relationship. To correct for this, we employ a sibling fixed effects model within the GxE framework. The intention is to mitigate potentially unseen gene-gene interactions or gene-environment correlation (rGE). In theory, siblings are identical in 50% of genes; therefore, the use of sibling fixed effects eliminates ~50% of the unobserved genetic variation that could be associated with the candidate gene in the GxE study. The use of sibling fixed effects does not completely eliminate the possibility for gene-gene interactions; however, the use of sibling fixed effects should provide a viable check for potentially confounding GxG

² This finding has important policy implications related to the beneficial effects of breastfeeding. For example, if favorable variants of FADS2 differ across socio-economic status, then policies that uniformly increase breastfeeding across class would be associated with increased inequality in IQ between SES groups (Lundborg and Stenberg 2010).

or rGE effects. Additionally, the use of sibling fixed effects allows for greater control of unobserved, shared family environments (e.g., parental education, family income, etc).³

This sibling strategy has been included in very few papers in the literature. Conley and Rauscher (2013; CR) show that previous GxE findings are indeed suspect when controlling for a shared familial environment. CR's main empirical strategy involves testing previous GxE findings in the presence of sibling fixed effects for both DZ and MZ twins. The use of MZ twins is used to eliminate any potential genetic differences between siblings, where the effect of the environmental exposure is measured across twin pairs containing differences in their genetic endowment. The use of MZ twins, while controlling for potential GxG effects within pairs, does not fully control for environmental differences between sibling pairs. To correct for this CR explore sibling fixed effects within DZ twins. The DZ analysis is similar to the current work, where DZ twins are as genetically similar as non-twin siblings. However, CR's baseline findings (i.e. before the use of sibling fixed effects) do not replicate or extend those in the literature, and are thus still preliminary.⁴

In this paper, we extend the GxE approach of Caspi et al. by using sibling comparisons from the Wisconsin Longitudinal Study. In particular, we examine the potential interactive effects between early favorable nutrition status, as measured by birth weight, and variation in the *FADS2* genotype in predicting young adult IQ. The main finding is that, while our baseline estimations are successful—we find similar results as the previous literature that genotype moderates the impact of early nutrition on later IQ—employing sibling comparisons shows the results and framework to be fragile to omitted family-level variables. The example has wider implications for the practice of investigating gene-environment interactions when the environmental exposure is not exogenous, and robust measures of the genome are not controlled in the analysis, and suggests the need to incorporate research designs from economics to other disciplines.

1.1 Gene-Environment Interaction and Early Nutrition Environment

A growing body of research is concerned with the development (i.e., natural selection) of differential genetic responses to particular environments. Foremost among these is the differential susceptibility hypothesis, which is more popularly known as “orchids and dandelions” (Belsky 2005). The main idea is

³ Our use of sibling fixed effects is not intended to eliminate all endogeneity in estimating the effect of gene-environment interactions. Rather, we seek to explore the robustness of these effects when accounting for time-invariant factors within families. The potential for bias in estimation remains as our approach (i) does not account for within-family time-varying factors and (ii) leaves open the possibility for unobserved differences between sibling genotypes being correlated with the candidate gene, environment, or outcome of interest.

⁴ Although Martin et al. (2011) uses a sibling/twin sample to attempt to replicate the Caspi findings, they do not use sibling fixed effects.

that some individuals (orchids) are more sensitive to environmental cues, thriving in good environments while struggling in harmful environments. Other individuals (dandelions) are relatively uninfluenced by variation in the environment, achieving the same outcomes regardless of the environment in which they are placed. In other words, the association of a genetic variant to an outcome is dependent upon the environment in which this variant is placed, with some variants being sensitive (orchids) while others are robust (dandelions). This hypothesis provides a reason as to why “harmful” genetic variants have persisted into contemporary times.⁵

Considering the differential susceptibility hypothesis, the use of breastfeeding as an environmental difference is problematic. The main argument by Caspi et al. is that certain variants of the *FADS2* gene interact with variation in breastfeeding to produce differential IQ scores. This implies that specific genetic variants were advantageous in specific breastfeeding environments. Breastfeeding, however, has remained constant for the vast majority of human history with the development of infant formulas occurring and becoming widespread in the second half of the 20th century (Castilho and Barros 2010). Given that this environmental difference is a relatively recent occurrence, it may be unlikely that genetic variants have been selected in response to the nutrition environment provided by breastfeeding. In other words, variation in *FADS2* is likely not strictly tied to differences in breastfeeding. This implies that the variants of *FADS2* may have differential effects for more general measures of the early nutrition environment. With this idea in mind, we use birth weight, not breastfeeding, as our primary measure for the early nutrition environment.⁶

The interaction between breastfeeding and *FADS2* is due to long-chained polyunsaturated fatty acids (LCPUFAs). The main LCPUFAs under consideration are docosahexaenoic acid (DHA) and arachidonic acid (AA), which are associated with early cognitive development (McCann and Ames 2005). Breast milk contains high levels of these LCPUFAs, and the *FADS2* gene is associated with extracting LCPUFAs from the diet. This gives rise to the hypothesis tested in Caspi et al. As stated earlier, we use birth weight as our environment in place of breastfeeding.⁷ Given that *FADS2* is associated with extracting LCPUFAs from the diet, birth weight may also have an interactive effect with *FADS2* variants.

⁵ Where harmful in this case is determined by interaction with a particular environment (see e.g., Caspi et al. 2002, Caspi et al. 2003, Guo et al. 2008).

⁶ The WLS data does not contain information on breastfeeding. As will be described in Sec. 2.1, the WLS data are primarily on Wisconsin high school seniors in 1957. Given that the use of infant formulas became widespread by the 1940s, it is safe to assume that the vast majority of our sample was breastfed.

⁷ A large body of literature explores the impacts of birth weight across and within families (see e.g., Conley and Bennett 2000, Conley et al. 2003, Hack et al. 2002),

Birth weight is a proxy for the early nutritional environment, and those infants who were exposed to better diets, including access to LCPUFAs, could have the same interactive effects with *FADS2* as breastfeeding.⁸ Furthermore, a number of studies have noted a correlation between LCPUFAs and birth weight (Leaf et al. 1992, Muthayya et al. 2009, Ramakrishnan et al. 2010). While the use of birth weight instead of breastfeeding does not allow us to replicate the findings of Caspi et al., our approach aims to extend the previous finding for a broader measure of early nutritional status that, based on the arguments above, could plausibly be hypothesized to interact with *FADS2* in a manner similar to that proposed by Caspi et al.

2. Data and Empirical Strategy

2.1 Data

The Wisconsin Longitudinal Study (WLS) is a randomly selected sample that is comprised of one-third of the 1957 high school graduates from Wisconsin. Information on graduates for a large number of individual and family characteristics were collected in 1957, 1964, 1975, 1992, and 2003, while information on selected siblings of the graduates began in 1977 with additional data collected in 1993 and 2004. For graduates, data were collected for 10,317 individuals, while for selected siblings, data were collected for 6,619 individuals.⁹

Our outcome variable of interest is IQ, which is mapped from a Henmon-Nelson test score from both the graduate's and sibling's junior year in high school. The independent variables of interest are birth weight, the allele frequency for two SNPs in the *FADS2* gene—rs174575 and rs1535, and the interaction (GxE) between these two variables. Following Caspi, important covariates include gender, race, age, mother's education, father's education, and a family-level score for socio-economic status in 1957. Summary statistics for our outcome variable and regressors of interest are given in Table 1.

⁸ The use of birth weight as a measure for early childhood nutrition is supported by Conley et al. (2006), who use birth weight as a proxy for differential nutrition environments within twins.

⁹ The collected data for siblings differ in regards to the collection of other variables and other waves. In the initial 1977 wave, selected siblings were randomly chosen from respondents if (a) the graduate had a sibling, (b) the graduate's sibling is between ages 19 and 66 (in 1977), and (c) preliminary data (from the 1975 graduate wave) of the selected sibling were known (i.e., sex and position in family roster). For the 1977 wave, data were collected for 2,133 selected siblings. Following the 1992 (graduate) wave, data collection for all selected siblings was attempted. The order of data collection: 1) Siblings in the 1977 wave. 2) Additional selected siblings for which the associated graduate participated in the 1992 wave. n For the 1993 sibling wave, mail questionnaires were completed for 4,036 siblings, while 4,804 completed at least part of the phone questionnaire. For the 2004 wave, 4,271 siblings completed at least part of a phone interview. The increase associated with IQ is due to the available data being present in high school roles, which is not directly sampled by the WLS.

In order to measure the two genetic variants given by Caspi et al., we simply use an indicator for possessing two copies of the specified variant from Caspi et al. For SNP rs174575, we use an indicator for having two copies of the “C” variant, where having two copies of any one variant is referred to as homozygous.¹⁰ The same indicator is used for the “A” variant of SNP rs1353. To correct for any potential benefits of containing just one copy of each variant, we also create an additive measure for each SNP that counts the number of variants an individual possesses—e.g., 0 for no copies of the “C” variant, 1 for heterozygotes, and 2 for homozygotes of the “C” variant.¹¹

While we have IQ data for roughly 17,000 individuals, the number of observations is reduced through the collection of additional, necessary variables. First among these is birth weight. Birth weight is self-reported in the 2003 wave for graduates and the 2004 wave for the selected siblings. For graduates, 3,472 individuals were missing from the 2003 wave and 2,322 did not contain data for birth weight, giving birth weight data for 4,523 graduates. For siblings, 3,297 were missing from the 2004 wave and 1,226 are classified as inappropriate, giving data on birth weight for 2,096 siblings.¹² Additionally, 113 graduates and 40 siblings are missing data for at least one covariate. A further sample reduction occurred from availability of biomarker data, which was collected in 2007 for graduates and 2008 for siblings. Complete biomarker data for the two *FADS2* variants exist for 4,455 graduates and 2,442 siblings. Overlapping these data with birth weight and other covariates results in the loss 1,732 graduates and 1,212 siblings. Finally, to perform the sibling fixed effects analysis we reduce the sample to only sibling pairs that have complete information for both the graduate and the selected sibling. In other words, all individuals are dropped from the sample unless they contain all available data and have a sibling who contains all available data. This results in a further reduction of 2,975 individuals, giving a sibling sample composed of 978 individuals from 489 sibling pairs.¹³ Summary statistics for the differing samples are given in Table 1.

The primary cause for our sample truncations is due to a reduction in the sample for the 2003 (2004) wave. At this time, graduates were in their mid-sixties, and it’s plausible that surviving until the 2003 (2004) wave is correlated with our dependent variable, IQ. To correct for this, we construct two sets of attrition weights. The first accounts for the probability of living and participating in the

¹⁰ All individuals receive two copies of DNA: one from the mother and one from the father.

¹¹ Caspi et al. also find a favorable advantage for heterozygotes. We are unable, however, to replicate these results with the use of an indicator for heterozygous individuals.

¹² An inappropriate classification is given to siblings who either did not know their birth weight or refused to answer.

¹³ All graduates are linked with data for only one sibling.

2003(2004) wave of the WLS, for which we regress an indicator for individuals with birth weight and DNA data, our two variables of interest, on the individuals education level and IQ, a mortality indicator, and all base controls. The inverse of this predicted probability is then used as a weight in estimation, correcting for possible sample selection due to living to and participating in the 2003(2004) wave of the WLS. The second set of attrition weights considers selection from all individuals who contain DNA and birth weight data to sibling pairs with complete data. Therefore, we regress an indicator for being in our base sibling sample on IQ and all baseline controls while also controlling for the predicted probability of having both DNA and birth weight data. Again, the inverse of this probability is used as a weight in estimation.

One other cause for concern in using the WLS is the generalizability of the sample. While being equally composed of both men and women, the sibling-pair sample is composed entirely of peoples of European descent.¹⁴ Out of the 489 sibling none are black, and no other ethnicities are represented. For our purposes in reproducing the findings of Caspi et al., however, the narrow focus of our sample is not a problem, as the sample of Caspi is also mostly composed of European derived ethnicities.

2.2 Empirical Strategy

In an effort to extend the findings of Caspi, we use birth weight as an indicator of early childhood (including in utero) nutrition. This approach, however, does not allow for an identical comparison to the gene-environment interaction of Caspi. Before estimating our proposed gene-environment interaction model, we perform additional estimations to check for the main, not interactive, effects of birth weight and the two SNPs of the *FADS2* gene on IQ, as well as check for a potential correlation between our genetic measures and our environment, birth weight.

The remainder of our study design is twofold. Firstly, we will estimate our GxE findings, which can be seen as an extension of the estimates from Caspi et al. Secondly, we will estimate the effects of this proposed gene-environment interaction while conditioning on a shared sibling environment and genome.¹⁵ More formally, our study will consider the following estimating equation:

$$IQ_{ij} = \beta_0 + \beta_1 SNP_{ij} + \beta_2 BW_{ij} + \beta_3 SNP_{ij} \times BW_{ij} + \beta'_4 X_{ij} + \beta'_5 Z_j + u_{ij} \quad (1)$$

¹⁴ This is not due to the sample truncations, but rather, the demographic composition of Wisconsin in the late 1950s.

¹⁵ Siblings share 50% of the genome passed from parents.

where β_3 is our coefficient of interest and represents the effect of gene-environment interaction for i individuals in j sibling pairs. OLS estimation of Equation (1) performs a similar estimate to the main findings of Caspi et al., and the use of sibling pairs will allow us to control for unobserved, sibling-shared omitted variables.¹⁶

The potential for omitted variable bias can be seen in the composition of the error term, u_{ij} . In addition to a random component, the error term is composed of unseen sibling-shared variation, both genetic and environmental, as well as individual specific variation. This is shown by:

$$u_{ij} = s_j + g_{ij} + e_{ij} + \varepsilon_{ij} \quad (2)$$

where s_j represents the unobserved, sibling-shared genetic and environmental effects. The use of sibling fixed effects would correct for potential omitted variable bias due to the unobserved, sibling-shared effects. In other words, if $cov(SNP_{ij} \times BW_{ij}, u_{ij}) \neq 0$ due to the $cov(SNP_{ij} \times BW_{ij}, s_j)$, then the use of a sibling fixed effects model will eliminate this potential source of bias.¹⁷ Correcting for this bias will give a more accurate effect of the GxE interaction resulting from variants of the *FADS2* gene and early childhood nutrition.

3. Results

3.1 Baseline Estimation

The structure of our tables will follow the following form. Column (1) performs OLS estimation of Equation (1) with the largest possible sample. Column (2) repeats the estimation of column (1) but uses the sibling pair sample. Column (3) weights the estimation of column (2) by the inverse of the probability of being in the sibling pair sample. Finally, column (4) controls for sibling fixed effects. All tables are broken into two panels with panel A using the rs174575 SNP and panel B using the rs1535 SNP.

Table 2 begins by exploring the main effects of the CC genotype for SNP rs174575, the AA genotype for SNP rs1535, and birth weight. All estimations of Table 2 control for gender, race, age, birth

¹⁶ The use of a sibling sample may also reduce bias in estimation itself, as the number of shared characteristics between treated (favorable homozygotes for *FADS2*) and untreated increases.

¹⁷ The possibility of unobserved, individual specific genomic or environmental bias remains, however.

order, family size, mother's education, father's education, and socio-economic status of the family in 1957 with standard errors clustered at the family level.¹⁸ Using as large as possible a sample in column (1) shows that each variant of *FADS2*—rs174575 and rs1353—is insignificantly associated with high school IQ scores, while birth weight has a positive and significant association, with roughly each standard deviation increase in birth weight being associated with a one point increase in IQ. Column (2) restricts the sample to sibling pairs, leading to no major change in the associations of column (1); however, the magnitude of the coefficient of standardized birth weight increases from 0.88 to 1.55 while remaining significant at the 1% level. This effect is consistent in both Panel A and Panel B, which estimate the effect of the rs174575 locus and the rs1353 locus, respectively. The estimation of column (3) weights the OLS estimation of column (2) by the inverse probability of being in the sibling-pair sample. This weighting causes no consequential change in the magnitude or significance of the coefficient of birth weight or each measure of the *FADS2* gene: the effect of birth weight remains positive and significant, while *FADS2* has no direct effect on IQ. Finally, in column (4) we control for sibling level fixed effects. The inclusion of sibling fixed effects does not cause a consequential change in the coefficient of either *FADS2* or birth weight. Each variant of *FADS2* has an insignificant association with IQ, while the effect of birth weight is consistent with previous findings.

The estimates of Table 2 support previous findings that birth weight is indeed a significant source of variation in later-life IQ (Black et al. 2007), while each locus of *FADS2* has no direct effect. Given the consistency of the coefficient of standardized birth weight, we have little reason to suspect this to be a spurious relationship. From Caspi et al., breastfeeding is associated with 5-6 point increase in IQ. From the estimations of Table 2, this large of an effect on IQ would be associated with a 3-4 standard deviation difference in birth weight. The 5-6 point difference in IQ found between being breastfed and not should be viewed with caution. Breastfeeding is a choice made by a mother, and this choice may be associated with other choices that influence later life IQ (Fletcher 2011).¹⁹ In other words, the effect of breastfeeding may be biased by unobserved heterogeneity.²⁰ The early nutrition

¹⁸ When employing sibling fixed effects race, mother's education, father's education, and socio-economic status are omitted due to these being shared controls amongst siblings.

¹⁹ Birth weight is also influenced by choices of the mother during pregnancy. However, the use of sibling fixed effects should control for consistent choices across siblings. And given the consistency in the coefficient of birth weight, this is unlikely to be a source of bias.

²⁰ Caspi et al. do mention the possibility of bias estimation due to SES status and maternal education. Their analysis, however, is problematic. Firstly, their measure of SES status is imprecise, simply grouping individuals into one of three SES classifications. Secondly, Caspi et al. don't directly control for SES status in their estimations. Instead they argue that since variants of *FADS2* do not have a significant interaction with SES in explaining IQ that

environment may also have a larger impact on IQ in earlier years. The IQ measure of Caspi et al. is found by averaging IQ from 7-13 years of age, a period significantly earlier than that measured in the current work.

Table 3 reproduces the estimation strategy of Table 2 but replaces IQ with standardized birth weight. The purpose of Table 3 is to show that no significant association exists between our gene and environment. A significant association between the gene and the environment would cause the gene-environment interaction to be suspect. It may not be the interaction, but rather the gene that is causing both selection into a particular environment and the outcome of interest. This concern is reduced by the estimates of Table 3, which finds an insignificant relationship between each SNP of *FADS2* and birth weight, our proxy for the early nutrition environment.

3.2 Gene x Environment

The structure of our tables will follow the following form. Column (1) performs OLS estimation of Equation (1) with the largest possible sample. Column (2) repeats the estimation of column (1) but uses attrition weights for the probability of having both DNA and birth weight data. Column (3) performs OLS estimation of our interactive model for our base sibling sample, while column (4) weights the estimation of column (3) by the inverse of the probability of being in the sibling pair sample. Finally, column (5) controls for sibling fixed effects. The primary comparison we wish to make is between columns (3) and (5) of Table 4, where the estimation of column (3) excludes sibling fixed effects leading to possible bias in estimation, while the estimates of column (5) include unobserved family-level controls.

Figure 1 shows the differential effect of birth weight from the variants of each *FADS2* SNP. Panel A of Figure 1 plots the effect of birth weight for those with two copies of C-allele of SNP rs174575 versus those without two copies of the C-allele. As is shown, homozygotes of the C-allele have a responsive, positive association between birth weight and IQ, while those without two copies of the C-allele exhibit no association between birth weight and IQ. The same is also true for A-allele homozygotes of SNP rs1535 in Panel B. This differential association with birth weight exhibits a textbook example for the differential susceptibility hypothesis (Belsky 2005), in which some individuals are sensitive to the environment—homozygotes of the C-allele for SNP rs174575 and homozygotes of the A-allele for SNP rs1535—while others are robust.

it is unlikely that SES status is accounting for the effects of breastfeeding. The variation in breastfeeding behavior within each SES class introduces measurement error into the estimation, leading to the insignificant interaction between *FADS2* and SES. To accurately measure the effect of breastfeeding, SES status should be included into the estimation.

The gene-environment interaction is tested in Table 4. Table 4 repeats the estimations of Table 2 while including the GxE interaction into all regressions. The results of Table 4 are fairly stark. A significant GxE interaction is found in all estimations until the inclusion of sibling fixed effects. When sibling fixed effects are included into the estimation the coefficient of the GxE interaction is reduced by roughly half and becomes statistically insignificant. One possible cause for this large reduction in the GxE coefficient is that unobserved, time-invariant heterogeneity shared between siblings is no longer a source of bias. A major candidate for this unobserved heterogeneity is the unmeasured genetic variation shared between siblings.²¹ The results of Table 4, particularly the contrast in the GxE coefficient between columns (5) and (6), do indicate that the significant GxE interaction between *FADS2* and birth weight is sensitive to controlling for time-invariant within-family heterogeneity.

While the point estimate of the coefficient on the interaction between birth weight and *FADS2* is reduced by roughly half from the inclusion of sibling fixed effects, we cannot rule out that the magnitude is inconsistent between the simple OLS of column (3) and the within-family specification of column (5). The reduction in precision from the within family estimation may be the cause of the insignificant coefficient of interest in column (5). As seen in Table 1, the within family variation is roughly half that for the sample. And we cannot rule out that this reduction in variation, which leads to more imprecise estimation within families, is the driving factor in the loss of significance seen in the estimates of column (5). Furthermore, given the self-reported measure of birth weight and the corresponding likely error in measurement, the use of the within family estimator may be exacerbating the attenuation of the coefficient associated with classical measurement error (Griliches 1979, Bound and Solon 1999).

As a check for the findings of Table 4, Table 5 re-estimates the findings of Table 4 with an additive measure for the sensitive variant of each *FADS2* SNP. The additive measure is intending to capture degrees of difference in the number of sensitive variants, and also controls for any beneficial effects of heterozygotes versus homozygotes for the robust alleles.²² The results of Table 5 are consistent with those of Table 4: A significant GxE interaction exists in standard OLS estimation, but this effect is substantially reduced by the inclusion of sibling fixed effects, resulting in an insignificant coefficient for the GxE interaction.

²¹ In addition to the shared genotype between siblings, familial environments which are constant over time and siblings are also controlled for through the use of sibling fixed effects.

²² As mentioned in Sec. 2.1 and footnote 5, Caspi et al. find a significant interaction between heterozygotes and breastfeeding.

The estimates of Tables 4 and 5 suggest that the previous findings of Caspi et al., and similar results from papers that are unable to control for shared family factors, should be taken cautiously. For one, the estimated effect of breastfeeding may be biased upwards due to inadequately controlling for SES status, which is positively correlated with breastfeeding and IQ. Furthermore, the inability by Caspi et al. to control for unobserved genetic heterogeneity leaves open the possibility that the interaction between *FADS2* and breastfeeding is the spurious byproduct of an unseen gene-gene interaction. The use of sibling fixed effects allows us to partially control for unobserved genetic differences. Doing so, results in an insignificant effect for the previously robust interaction between *FADS2* and the early nutrition environment and calls into consideration the true nature of this relationship.

4. Conclusion

This paper finds evidence that the interaction between the *FADS2* gene and early life nutrition may have no effect on later-life IQ. Rather, the baseline GxE associations we find may be the product of unobserved, familial characteristics. In order to identify an interaction between a single environment and a single allele, other observed and unobserved differences in the genome and the environment must be accounted for. While not being able to control for all genetic and environmental differences, sibling fixed effects provides an important robustness check for the influence of unobserved, sibling-shared genes and environments. Along with Conley and Rauscher (2013), the current work calls into question many previously found GxE associations.

In summary, we argue that birth weight is a viable proxy for the early nutrition environment and could interact with *FADS2* in a similar manner to breast feeding. The GxE interaction between *FADS2* and birth weight is indeed shown to have a positive and statistically significant effect on later-life IQ when we employ the typical specification used in this literature, a finding that echoes the interaction effect of *FADS2* and breastfeeding from Caspi et al. Importantly, the statistical significance of this GxE interaction, however, dissipates with the inclusion of sibling fixed effects. Thus, our findings may also question previous results linking early nutrition and *FADS2* genotype with IQ. More broadly, our example has wider implications for the practice of investigating gene-environment interactions when the environmental exposure is not exogenous and robust measures of the genome are not controlled in the analysis, and suggests that incorporating research designs from economics and other social sciences into the health and biological sciences can lead to fewer false positive results. This is important both in

enhancing the robustness of research on gene-environment interactions and also so that policies are not created based on fragile findings from these literatures.

5. References

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