Revisiting the Children of Twins: Can They Be Used to Resolve the Environmental Effects of Dyadic Parental Treatment on Child Behavior?

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The children of twins (COT) design has been proposed as an alternative to the adoption study to resolve the direct effects of parental treatment from secondary parent–child association due to genetic factors. The basic analytical approach compares the parent–offspring correlation with the correlation between children and the monozygotic (MZ) twins of their parents. We show that a significant difference between these correlations does not imply direct environmental causality when the measured parental treatment in question is dyadic, that is, influenced by both parents even when mating is random. Nongenetic causal effects yield very similar patterns of correlation to secondary genetic effects on dyadic treatment variables. The fact that many candidate environments, such as parental divorce, are dyadic gives reason to question the interpretation of their correlations with behavior in the children of twins.

There has been growing interest in using the children of twins (COT) to help resolve the direct nongenetic causal effects of parental treatment on child behavior from secondary association due to the shared genetic characteristics of parents and children (e.g., D'Onofrio, in press; D'Onofrio et al., 2003; Rutter et al., 2001; Silberg & Eaves, 2004). Several models and applications have been offered for the kinships of twins (e.g., D'Onofrio et al., 2003; Eaves, 1982; Eaves et al. 1999; Haley et al.,1981; Heath et al.,1985; Lake et al., 2000; Maes et al., 1997; Nance & Corey, 1976; Silberg & Eaves, 2004; Truett et al., 1994). These models were focused primarily on parental traits such as schizophrenia (Gottesman & Bertelsen, 1989) measured on individual parents. The unique value of the COT lies in its potential to resolve the direct causal effects of individual parental characteristics from secondary noncausal association due to genetic factors. Recent applications, however, have used the COT design to analyze the effects of exposure to dyadic parental treatments, such as divorce, that aggregate the behavior of individual parents (e.g., D'Onofrio et al., in press). Such analyses rely on the intuitively appealing extension of the basic logic of the COT design, developed originally for the analysis of family resemblance for measures of individual parents and children, to the case of dyadic variables that aggregate the behavior of both parents.

Upon closer scrutiny with the aid of an explicit structural model for parent–offspring transmission, it turns out that the usual causal interpretation of COT does not extend easily to dyadic variables such as divorce and marital conflict. Dyadic treatment data may yield COT data superficially consistent with a causal hypothesis when, in fact, there is no direct path from treatment to outcome. Furthermore, a model that makes explicit allowance for the dyadic nature of the environment may yield parameter estimates that are so highly correlated as to cast serious doubt on the ability of COT to resolve causal from secondary genetic associations of dyadic parental treatment and childhood outcome with current sample sizes. Even when mating is random, it appears that the COT design yields more ambiguous conclusions with dyadic treatments than has been appreciated thus far.

Model for Effects of Dyadic Treatment

Figure 1 shows the essential features of the model for the effects of parental treatment, T, on behavior in the offspring of twins, P. P_1 and P_2 represent the outcome phenotypes in pairs of cousins derived from a pair of twin parents. The offspring of monozygotic (MZ) pairs are biologically half siblings (Haley et al., 1981; Nance & Corey, 1976) and the offspring of dizygotic (DZ) pairs are biologically first cousins. The measured family environments of the offspring are represented by T_1 and T_2. Random, residual effects on the offspring are the latent variables E_1 and E_2. The

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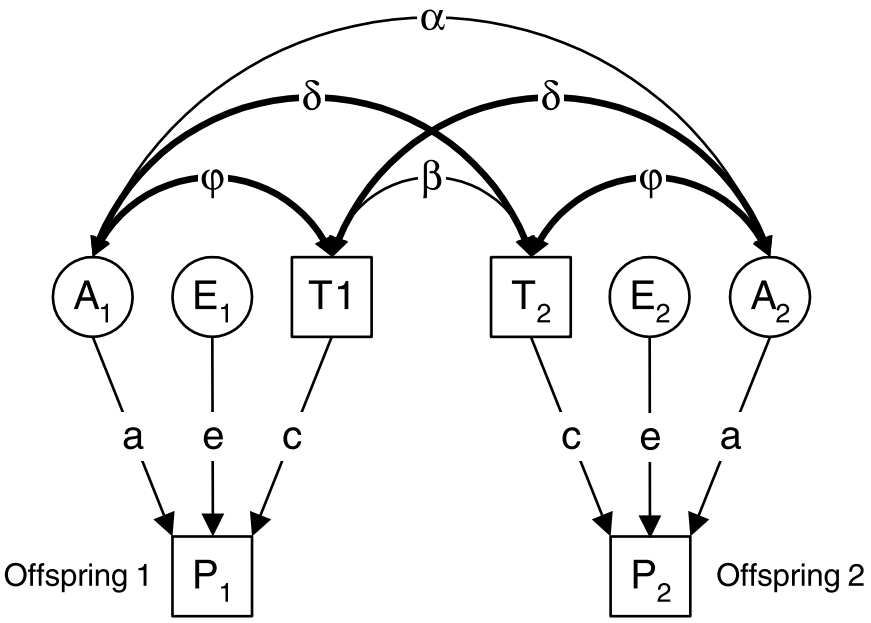

Figure 1

Path model for relationships between genes (A1 and A2), treatment (T1 and T2), and phenotype (P1 and P2), in the offspring of pairs of adult twins.

Note: The pathways highlighted in bold arrows are expected to differ when the treatment is dyadic (i.e., the aggregate of behavior of both parents) because the passive genotype-environment within nuclear families exceeds the avuncular genotype–environmental correlation ($\phi > \delta$).

latent variables, A_1 and A_2, are the genetic effects on the offspring. The variables are assumed to be standardized. The paths *a*, *c* and *e* represent the (linear) effects of A, T and E on offspring behavior. Residual genetic and shared environmental effects on children are not included in the diagram and do not affect the basic conclusions of this discussion.

The fact that offspring derive both genes and environment from their parents implies that there will be a passive genotype–environment correlation, ϕ, between the effects of genes and treatment within offspring. Insofar as the behavior of twin parents is correlated, there will be a correlation, β, between the treatments of cousin offspring. Similarly, the genetic effects of cousins are expected to show correlation, α, depending on the zygosity of parents. Finally, the biological relationship between twin parents produces a passive genotype–environment correlation, δ, between the offspring genotype and the treatment of his/her cousin.

The model in Figure 1 is similar to the familiar ACE model for twins (e.g., Neale & Cardon, 1992). However, the latent shared environmental variable, C, of the ACE model is replaced by a measured (dyadic) variable T; the genetic correlation between pairs, α, is that appropriate for cousins related through MZ or DZ twins ($^1/_4$ and $^1/_8$ respectively under random mating) and the correlation between shared environmental treatments, β, will depend on the zygosity of the twin parents A further difference between Figure 1 and the ACE model is the inclusion of correlations between the genotypes (A) and treatments (T) of offspring.

Although there are many biological and social relationships in the kinships of twins (Eaves et al., 1999; Maes et al., 1997; Truett et al., 1994), analysis of the causal relationships between parental treatment and child outcome depends critically on comparing the correlations between phenotype and treatment within nuclear families ($r_{T1,\,P1}$ and $r_{T2,\,P2}$) with the corresponding correlations between offspring phenotype and treatment (e.g., divorce) assessed in the twins, especially monozygotic twins, of parents ($r_{T1,\,P2}$ and $r_{T2,\,P1}$). In the usual application of the COT design, treatment is assumed to be assessed independently in both parents (e.g., paternal and maternal depression). In the application to dyadic treatments such as divorce, this is not the case since the breakdown of marriage may depend upon genetic and environmental influences on the behavior of both parents. It turns out that this difference is critical for the analysis of the COT design, even in randomly mating populations.

Applying the rules of path analysis, the model in Figure 1 yields:

$$r_{T1,\,P1} = r_{T2,\,P2} = c + a\phi$$

and

$$r_{T1,\,P2} = r_{T2,\,P1} = c\beta + a\delta.$$

If the path *c* is significant, then it may be inferred that T has a significant direct and causal effect on P. However, since the correlation between treatment and outcome within nuclear families also has a genetic

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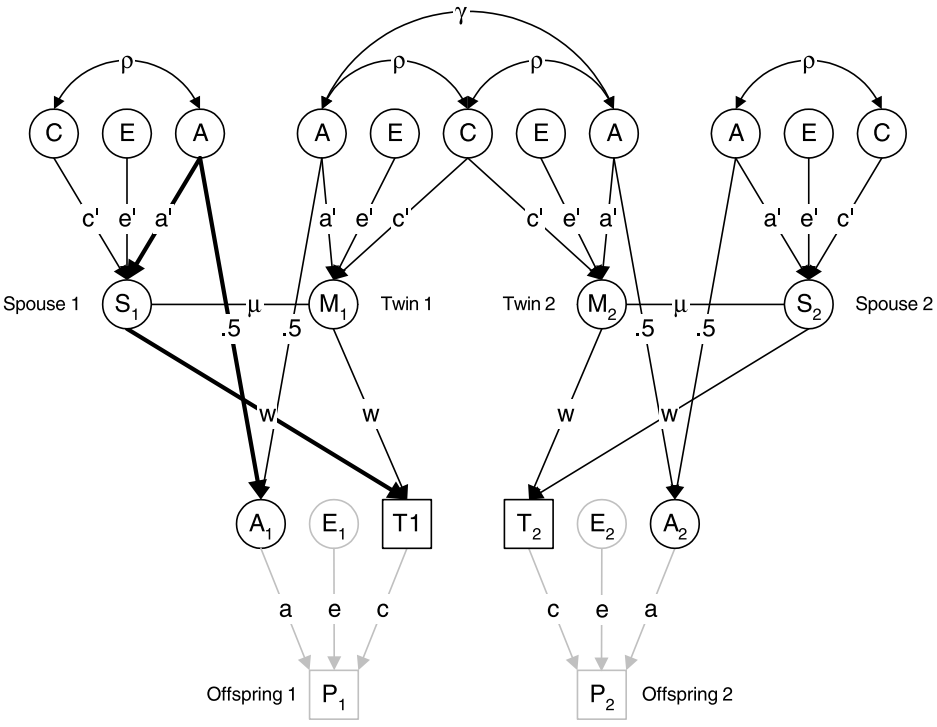

Figure 2

Model for contributions of twins and spouses to genetic and treatment differences in the children of twins.

Note: The genotype-environmental pathway represented by bold arrows, from offspring genotype (A1) through spouse's phenotype (S1) to measured environment of offspring (T1) contributes to the correlation, ϕ, between offspring genotype (A1) and dyadic parental treatment (T1) but does not contribute to the correlation, δ, between A1 and treatment in the co-twin's family (T2).

component, $a\phi$, the magnitude of the correlation is biased by the effect of parental genotype on treatment. It is this bias that the COT design attempts to eliminate by comparing $r_{T1,\,P1}$ and $r_{T2,\,P2}$ with $r_{T1,\,P2}$ and $r_{T2,\,P1}$. The weight of the argument rests on the assumption that the environments in the families of MZ co-twins are not the same as those in parents ($\beta < 1$) but the twins' genotypes are identical. Thus, any significant excess of $r_{T1,\,P1}$ and $r_{T2,\,P2}$ over $r_{T1,\,P2}$ and $r_{T2,\,P1}$ justifies the inference that c is greater than 0, or that T exercises a causal effect on P.

The above expectations show that the critical assumption underlying the inference of causation is that the genotype–environment correlations, ϕ and δ, are identical in the above formulae (see pathways represented by bold arrows in Figure 1). Although it is acknowledged that this is not the case in the presence of assortative mating, the assumption is not justified even under random mating when the environmental treatment depends on the aggregate behavior of both parents, such as parental marital conflict. This assumption may not be true even for individual-specific assessments such as maternal psychopathology if there is marital interaction (Heath, 1987).

The genotype–environment correlations may be resolved in terms of genetic and environmental effects on parental treatment and their transmission to offspring. The initial model of Figure 1 is extended back to the previous generation in order to evaluate the difference between ϕ and δ. Figure 2 shows a simplified path model for the transmission of A and T between twins and their children.

The diagram assumes that T in nuclear families depends on the phenotypes of twin mothers, M, and their unrelated spouses, S. For simplicity it is assumed that the paths, w, from M and S to T are the same. Thus, T is a dyadic variable caused equally by both parents. M and S both depend on the three sources of variance typically assumed to contribute to phenotypic differences and resolved in twin studies as additive genetic effects, A, shared environmental effects, C, and unique environmental effects, E. It is assumed, again for simplicity, that the paths, *a'*, *c'* and *e'*, from A, C and E respectively to mothers' and fathers' phenotypes are the same in both sexes. The correlation between genotypes of twins is γ, and the passive genotype environment correlation between A and C produced by the grand-parental generation is ρ. The intergenerational path from additive genetic effects in parents to the additive genetic effects in children is known from genetic theory to be 1/2. In MZ twins the genetic correlation, γ, is unity. When mating is random, γ is 1/2 in DZ twins. If there is assortative mating γ is greater than 1/2 by a factor that depends on the heritability

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and the correlation between mates (Fisher, 1918). Given the possible importance of assortative mating, we introduce a correlation, μ, between spouses' phenotypes. Assortment is represented by the 'copath' notation of Cloninger (1980, see also Fulker, 1988). This notation simplifies both the diagram and the derivation of correlations by representing implicitly rather than explicitly the many correlations between genetic and environmental effects of spouses. Although the diagram assumes the twins are female, the sexes of twins and spouses may be interchanged in this case without affecting the result, as all genetic and environmental effects are assumed to be the same in males and females for this simple illustration.

From Figure 2, we may then derive the correlations:

$\beta = w^2(1 + \mu)^2\, t$, where t is the correlation between twins' phenotypes, M_1 and M_2;

$\alpha = {}^1/_4\,[\gamma + 2\mu\,(a'+c'\rho)(a'\gamma + c'\rho) + \mu^2 t(a'+c'\rho)^2]$

$\phi = w(a'+c'\rho)(1+\mu)$ and

$\delta = {}^1/_2\, w[a'\gamma + c'\rho + \mu t(a'+c'\rho)](1+\mu)$

When mating is random $(\mu = 0)$ the correlations in families of MZ twins $(\gamma = 1)$ simplify to:

$\beta_{MZ} = w^2\, t_{MZ}$; $\alpha = {}^1/_4$; $\phi = w(a'+c'\rho)$ and $\delta = {}^1/_2\, w(a'+c'\rho)$

Implications for Interpretation of COT Data on Dyadic Treatment

Even when mating is random and there is no direct effect of parental treatment, T, on outcome, P, the critical passive genotype–environment correlation, ϕ, exceeds δ, yielding an excess of the treatment–outcome correlation in nuclear families compared with that between offspring and the MZ twins of their parents. The critical pathway, between the treatment of offspring (T_1) through the spouse's phenotype (S_1) to the offspring genotype (A_1), is shown with bold arrows in Figure 2. This pathway does not contribute to the equivalent avuncular relationship (e.g., between A_1 and T_2) when treatment is dyadic. Thus, when parental treatment is dyadic, a simple comparison of the offspring–treatment correlations (e.g., $r_{T1,\,P1}$ with $r_{T2,P1}$) or any statistical test that depends on this comparison, is likely to be misleading about the causal impact of parental treatment on outcome.

The reason is simple but relatively obscure unless the model is specified fully. The passive genotype–environment correlation in nuclear families depends on the genetic and environmental effects of *both parents* in the family when T is affected by both parents (i.e., T is dyadic). The corresponding correlation between cousins (even when they are offspring of identical twins) only reflects the effects of the *one twin parent* because the offspring receive their genes and treatments from different fathers.

Furthermore, if T only depends on the twin parent (e.g., mothers' smoking in pregnancy under simple assumptions) and there is no contribution of fathers to the treatment variable, then the path from spouses' phenotypes to treatment, w, will be set to zero, reducing ϕ to ${}^1/_2 w(a'+c'\rho)$ when mating is random, satisfying the condition for concluding that differences between correlations reflect only the direct causal effect of treatment on outcome. This is not the case for dyadic treatments insofar as they reflect contributions of both parents.

When mating is random and there is no genotype environmental correlation in the parental generation (i.e., $\rho = \mu = 0$) the correlations simplify to:

$r_{T1,\,P1} = r_{T2,\,P2} = c + a\phi = c + awa'$

and

$r_{T1,\,P2} = r_{T2,\,P1} = \beta c + a\delta$

which, in MZ twins, becomes:

$r_{T1,\,P2MZ} = r_{T2,\,P1MZ} = \beta_{MZ} c + a\delta_{MZ} = \beta_{MZ} c + {}^1/_2\, awa'$

and, in DZ twins:

$r_{T1,\,P2DZ} = r_{T2,\,P1DZ} = \beta_{DZ} c + a\delta_{DZ} = \beta_{DZ} c + {}^1/_4\, awa'$

That is, if the twin correlations, β_{MZ} and β_{DZ} are known for the parental treatment index, the three correlations can be expressed in terms of two parameters: the direct causal effect of the treatment on phenotype, c, and the partially confounded effects of the non-causal correlation between parental treatment and genotype, $z = awa'$.

An intuitively appealing test for the causal effect of treatment on outcome is the correlation between twin differences in treatment $(T_1–T_2)$ with the differences in outcome between the cousin offspring of MZ twins $(P_1–P_2)$. At first sight, this is an extension of the comparison of risk to disease in the children of twins discordant for a risk factor (e.g., Gottesman & Bertelsen, 1989). However, when the parental risk factor is dyadic, the direct causal effects of treatment are confounded with the indirect genetic effects of the spouses of twins. The latter are likely to be as great as the genetic effects of the twin parents when treatment is dyadic so cannot reasonably be ignored.

The covariance between the differences within twin pairs and (MZ) cousin pairs is

$r_{T1,\,P1} + r_{T2,\,P2} - r_{T1,\,P2MZ} - r_{T2,\,P1MZ} = 2(1-\beta_{MZ})c + a'wa$

when treatment is dyadic and mating is random.

That is, even when there are no direct effects of dyadic treatment, c, the covariance is greater than zero because of indirect genetic effects $(z = a'wa)$. When the measure of treatment depends only on one parent, the genetic bias disappears and the intuitive rationale is justified under random mating. Otherwise the test does not give any uniquely identified information about the causal association between dyadic treatment and outcome. It turns out that the expectations for the covariances for other relationships (unrelated and DZ twin pairs) yield very similar patterns (cf., e.g., D'Onofrio et al., in press, Figure 1) whether the effect of a dyadic parental treatment is causal and environmental or secondary and genetic. Thus, the covariance between cousin differences and the treatment by their DZ twin parents is expected to be $2(1-\beta_{DZ})c + 1{}^1/_2 a'wa = 2(1-\beta_{DZ})c + 1{}^1/_2 z$. For pairs of unrelated

Table 1

Expected Regression of Offspring Differences on Parent Differences for Monadic and Dyadic Treatments in Kinships Derived From Different Parental Relationships

Parental relationship	Statistic		
	Variance (T_1-T_2)	Covariance $[(T_1-T_2),(P_1-P_2)]$	Regression
	Monadic treatment		
Unrelated	1	$c + {}^1/_2 z$	$c + {}^1/_2 z$
DZ twins	$1-\beta_{DZ}$	$(1-\beta_{DZ})c + 1/4z$	$c + {}^1/_4 z/(1-\beta_{DZ})$
MZ twins	$1-\beta_{MZ}$	$(1-\beta_{MZ})c$	c
	Dyadic treatment		
Unrelated	1	c + z	c + z
DZ twins	$1-\beta_{DZ}$	$(1-\beta_{DZ})c + {}^3/_4 z$	$c + {}^3/_4 z/(1-\beta_{DZ})$
MZ twins	$1-\beta_{MZ}$	$(1-\beta_{MZ})c + {}^1/_2 z$	$c + {}^1/_2 z/(1-\beta_{MZ})$

Note: *c* is the path from treatment to child behavior; *z* is the composite secondary pathway through genetic effects of parents (a'wa). Variances and covariances of differences are multiplied by $^1/_2$ to simplify formulae.

Table 2

Expected Regressions of Offspring Differences on Dyadic Treatment Differences for Different Values of MZ and DZ Twin Correlations

Correlation		Regression		
MZ	DZ	Unrelated	DZ twins	MZ twins
.5	.25	c+z	c+z	c+z
.4	.2	c+z	c+.94z	c+.8z
.3	.15	c+z	c+.88z	c+.71z
.2	.1	c+z	c+.83z	c+.62z
0	0.	c+z	$c+{}^3/_4 z$	$c+{}^1/_2 z$

parents and differences between their children the covariance is expected to be $2(c + z)$. That is, in the case of mothers and fathers contributing equally to the measured home environment, COT cannot identify the passive correlation between dyadic parental treatment and offspring genotype even in the simplest case of random mating.

Tables 1 and 2 illustrate the expected regression of offspring differences on parent differences for unrelated, DZ and MZ parents. Table 1 gives the expected variances of parental differences ($\times {}^1/_2$), covariance of parent differences and offspring differences ($\times {}^1/_2$) and the regression coefficients for both monadic and dyadic treatment. The regression coefficients correspond to the difference expected between offspring outcomes for a unit difference in parental treatment.

Each expected regression (Table 2) involves the 'direct' (causal) effect of treatment, *c*, and the indirect (genetic) effect, *z*, defined above and in the Figures. The effect of *c* is a constant term regardless of whether the treatment is dyadic or not, and whether or not there is a 'backdoor' secondary genetic association. Furthermore, in the monadic case, the regression (case-control difference) in MZs is purely *c*. That is, when treatment depends only on one parent the COT design allows estimation of the direct effect of treatment uncontaminated by any secondary genetic association.

The situation is more confused in the dyadic case. Table 1 confirms the previous result that there is a genetic bias in the MZ pairs that is a function of *z* and the MZ correlation in treatment. Table 2 shows the expected size of the bias for different values of the MZ and DZ correlation for a dyadic treatment variable. If the MZ and DZ correlations in treatment are .5 and .25 respectively (the most extreme case of genetic effects on treatment under random mating for a dyadic variable), the case-control differences are identical for all three relationships and it is impossible to distinguish *c* from *z*. Furthermore, the COT design does not reduce the difference between cases and controls as a function of relationship.

At the other end of the scale of twin resemblance, when the twin correlations are both zero, there is a singularity because if the MZ treatment correlation is zero, then there also cannot be any indirect genetic effect. That means, when the twin correlation is zero all the control differences are expected to be the same also. So, when treatment is completely genetic or not at all genetic the case-control differences for all three parental relationships are the same across rows of the table, independently of the size of the 'genetic bias' in the case of genetic treatment differences.

Table 2 also provides the coefficients of the genetic bias (*z*) when the MZ correlation lies between 0 and .5 and the DZ correlation is half that of MZs. An MZ correlation of .3 with a DZ correlation of .15 (similar in magnitude to the correlations reported for divorce), yields case-control differences: unrelated parents = $c + z$ (as always); DZ = $c + 0.88z$; MZ = $c + 0.71z$.

If treatment is dyadic, then the absence of any MZ case-control difference would imply $c = z = 0$, which is inconsistent with any significant difference between unrelated cases and controls. A zero MZ difference alongside a large case-control or DZ difference, therefore, implies that the data are internally inconsistent with what is expected under the dyadic model, because 71% of the genetic bias goes into the MZ difference even when there is no direct treatment effect. Thus, the MZ difference should not differ excessively from the case control difference or the DZ difference. Otherwise, a true MZ case-control difference of zero predicts, within sampling error, a zero difference for the unrelated case-controls also. If there is an indirect effect and no direct treatment effect, the MZ difference should still not be zero and only about 30% smaller than the unrelated case-control difference.

Implications for Power

Application of COT to dyadic treatments imposes further constraints on the twin correlations because

variation in both parents contributes to differences in treatment. Thus, even if the parental treatment is completely 'genetic' in the sense that it depends on completely heritable aspects of parental phenotypes, the MZ correlation in treatment is expected to be only $\beta_{MZ} = {}^{1}/_{2}$ at most if mating is random and both spouses contribute equally to the dyadic treatment. That is, *genetic* variation in spouses creates *environmental* variation within MZ twin pairs.

Now in theory, at least when mating is random, the causal and noncausal association between dyadic parent treatment and offspring outcome can be resolved by solving the three equations for c and z once the twin correlations for treatment are known (cf. Silberg & Eaves, 2004). In practice, however, the coefficients of c (1, β_{MZ}, β_{DZ}) will usually be highly correlated with the coefficients of z (1, 1/2, 1/4) as in almost all cases we expect $1 > \beta_{MZ} > \beta_{DZ}$, especially if T is dyadic.

Writing $z = a'wa$, we consider simple least squares estimation of c and z from the three intergenerational correlations above, it may be shown that the variance of the estimate of c is proportional to:

$$V(c) = 1 + (1/2)^2 + (1/4)^2$$

The variance of z is proportional to:

$$V(z) = 1 + \beta^2_{MZ} + \beta^2_{DZ}$$

and the covariance of z and c proportional to:

$$W(z,c) = -(1 + 1/2\beta_{MZ} + 1/4\beta_{DZ})$$

whence the correlation between the estimates may be derived as a function of the twin correlations in treatment.

The large positive correlation between the coefficients leads to a large negative correlation between estimates of the direct and spurious effects of treatment, c and z. That is, the standard errors of estimates of c and z are likely to be enormously inflated in a model that properly reflects the ambiguity in the association between dyadic measures of parental treatment and offspring outcome. Clearly, in the special case $\beta_{MZ} = 1/2$ and $\beta_{DZ} = 1/4$ the correlation between the estimates is –1 and the direct and indirect effects cannot be resolved even with an infinite number of families. However, the situation is still very bad even in better cases. For example, if $\beta_{MZ} = \beta_{DZ} = 0$, the correlation between the estimates is –.873. When $\beta_{MZ} = \beta_{DZ} = 1$ it is –.882 and reaches –.946 if $\beta_{MZ} = 1$ and $\beta_{DZ} = 1/2$. Thus, over a wide range of values for the correlation in treatments provided by twins for their children, the correlation between estimates of c and z is large and negative. Even if the twin correlations in treatment were known precisely, the separation of the causal and noncausal association between dyadic measures in parents and offspring outcomes resembles very closely the separation of additive from dominance genetic effects in twins studies (see Eaves, 1972) for estimates of which the correlations are also large and negative and for which projected sample sizes for reliable resolution are very large indeed, probably beyond the scope of most COT samples. Put more simply, under random mating it is safe to infer causality only if the three correlations depart significantly from the proportions *1:* 1/2 *:* 1/4 expected under the hypothesis of purely *genetic* association for the case of dyadic variables. It is not sufficient, or even informative, to show that the parent–offspring correlation exceeds the avuncular correlation in kinships of MZ twins or that twin differences in treatment correlate with cousin differences in behavior.

Although it is possible to estimate c and $z = a'wa$ separately (i.e., by fitting each without the other), the resulting estimates are heavily biased by the missing parameter.

So, estimating c on the assumption that $z = 0$ leads to an estimate of c that is inflated by: $z(1 + 1/2\beta_{MZ} + 1/4\beta_{DZ}) / (1 + \beta^2_{MZ} + \beta^2_{DZ})$. Similarly, estimating $z = a'wa$ and ignoring c inflates z by $c(1 + 1/2\beta_{MZ} + 1/4\beta_{DZ}) / [1 + (1/2)^2 + (1/4)^2]$.

That is, the coefficients of the omitted parameters in the bias are very close to 1 in many cases suggesting that when either parameter is omitted from the model it emerges almost entirely as an upward bias in the other.

This combination of a large negative correlation between estimates of c and $a'wa$ when both are present, and the large bias when either is left out of the model, make the resolution of c and w in the dyadic case highly improbable with feasible sample sizes once the model is specified correctly. The preliminary simulations of Silberg and Eaves (2004) confirm that it is relatively easy to test for treatment–offspring association but much more difficult to resolve the causal and genetic sources of transmission. The current treatment makes the reason for this ambiguity, that is, the genetic pathway through spouses, far more explicit.

Discussion

The children of twins offer a significant conceptual and practical opportunity for separating the contributions of genes and treatment to the association between offspring and parental behavior. However, application of the COT design to dyadic treatment variables is unlikely to resolve direct causal associations from passive genotype–treatment correlation even when mating is random for risk factors in question unless sample sizes are large and parameter values are optimal. Insofar as the design was adopted to resolve the direct environmental effect on children from indirect genetic association, the results are expected to be disappointing. In the dyadic case, differences between cases and controls derived from unrelated parents and monozygotic twins are much more alike than might be expected intuitively. Furthermore, the differences between children of unrelated cases and controls are expected to comprise a quite similar and confounded mixture of direct and indirect effects.

Models that fail correctly to specify both the direct causal effects of treatment and the secondary indirect correlation due to passive genotype–environment correlation yield seriously biased estimates of the direct causal effect of dyadic parental treatment. Indeed, the high negative correlation expected between estimates of the two effects reduces the probability that they can be resolved with practicable sample sizes. The problem arises because the passive genotype–environment correlation in nuclear families depends on contributions of both parents whereas it depends only on one of the parents (who is the twin) in cousins because the spouses of twins (the other parent) contribute different genes and environments to their offspring. If it may be shown that only one parent affects the measured family environment, the problem disappears. In the case of divorce, for example, that would require that the twin correlations for divorce are zero in one sex. Empirically, this is not the case.

Using COT with dyadic parental variables fails to resolve critical ambiguity about the cause of parent–offspring association. This conclusion is disappointing if we look to this particular quantitative genetic design to help resolve alternative theories for the family origin of individual differences in behavior. It remains to be seen how far fitting an explicit structural model can solve the problem (see Silberg & Eaves, 2004). However, the very high negative correlation expected between parameter estimates implies that the direct causal effect of a dyadic treatment is unlikely to be resolved reliably from the indirect association due to genetic correlation between parents and children. Indeed, the simulations reported by Silberg and Eaves, where the resolution of genetic and cultural effects was considered the primary goal of COT, suggested that very large samples of COT families were likely to be required under otherwise quite favorable circumstances.

The problem persists as long as there are unmeasured characteristics of the individual twins and spouses that create variation in the dyadic treatment they provide for their children. How far the difficulty is ameliorated by measuring specific aspects of the individual parents (e.g., parental psychopathology) is not yet clear. Insofar as additional measurement can resolve contributions of mothers and fathers to the aggregate gene–environment correlation in their offspring, the COT design again reverts to the original form and models. Insofar as variation in treatment is dependent on latent, unmeasured, genetic and environmental influences on both parents, the basic ambiguity remains. Further extension of the basic structural model along the lines suggested by Meyer et al. (2000) for kinships of twins and their parents is needed to determine how far dyadic indices of the environment can be shown to mediate the impact of specific individual parental behavior. Analyses limited to the dyadic treatment itself do not accomplish this goal.

Although the problem may be critical for treatments that are inherently dyadic, our discussion also counsels caution in using the children of twins to explore other treatment variables that could depend on the phenotypes of both parents. For example, maternal depression may be influenced by paternal alcoholism. Thus even measures made on parents separately may still have some of the characteristics of dyadic variables for the purposes of detecting nongenetic transmission from parent to child. Careful examination of the patterns of association between variables in twins and their spouses should be undertaken as a necessary precursor to any interpretation of COT data in strictly causal terms. Although the COT design offers great advantages, it is no panacea and should be interpreted with the utmost caution unless the individual contributions of both parents to the family environment can be resolved. Specifically, the close confounding of direct causal effects with secondary genetic association even within the offspring of MZ twins, makes it less clear how much COT adds to classical studies of parent–offspring resemblance for characterizing the environmental impact of dyadic measures.

Although the basic result of this paper may seem painfully obvious with hindsight, we are not aware that the problem has been stated explicitly or its implications appreciated. Further theoretical and model-based data analytical studies are needed to establish the limits of the COT design for analyzing the impact of dyadic treatments such as parental divorce and marital conflict. Our theoretical analysis implies it is premature to claim that COT markedly reduces ambiguity about the causal relationships between dyadic parental treatment and offspring behavior.

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References

Cloninger, C. R. (1980). Interpretation of intrinsic and extrinsic structural relations by path analysis: Theory and applications to assortative mating. *Genetical Research, 36*, 133–145.

D'Onofrio, B. M. (in press). The children of twins design. In B. Everitt & D. Howell (Eds.), *Encyclopedia of behavior statistics*. New York: Wiley.

D'Onofrio, B., Turkheimer, E., Emery, R., Slutske, W., Heath, A., Madden, P., & Martin, N. (in press). A genetically informed study of marital instability and its association with offspring psychopathology. *Journal of Abnormal Psychology*.

D'Onofrio, B. M., Turkheimer, E. N., Eaves, L. J., Corey, L. A., Berg, K., Solaas, M. H., & Emery, R. E. (2003). The role of the children of twins design in elucidating causal relations between parent characteristics and child outcomes. *Journal of Child Psychology and Psychiatry, and Allied Disciplines, 44*, 1130–1144.

Eaves, L. J. (1972). Computer simulation of sample size and experimental design in human psychogenetics. *Psychological Bulletin, 77,* 144–152.

Eaves, L. J. (1982). The utility of twins. In V. E. Anderson, W. A. Hauser, J. K. Penry, & C. F. Sing (Eds.), *Genetic basis of the epilepsies* (pp. 249–276). New York: Raven Press.

Eaves, L. J., Heath, A. C., Martin, N. G., Maes, H. H., Neale, M. C., Kendler, K. S., Kirk K. M., & Corey, L. A. (1999). Comparing the biological and cultural inheritance of personality and social attitudes in the Virginia 30 000 study of twins and their relatives. *Twin Research, 2*, 62–80.

Fisher, R. A. (1918). The correlation between relatives on the supposition of Mendelian inheritance. *Transactions of the Royal Society of Edinburgh, 52,* 399–433.

Fulker, D. W. (1988). Genetic and cultural transmission in human behavior. In B. S. Weir, E. J. Eisen, M. M. Goodman, & G. Namkoong (Eds.), *Proceedings of the Second International Conference on Quantitative Genetics* (pp. 318–340). Sunderland, MA: Sinauer.

Gottesman, I. I., & Bertelsen, A. (1989). Confirming unexpressed genotypes for schizophrenia. Risks in the offspring of Fischer's Danish identical and fraternal discordant twins. *Archives of General Psychiatry, 46,* 867–872.

Haley, C. S., Jinks, J. L., & Last, K. (1981). The monozygotic twin half-sib method for analysing maternal effects and sex-linkage in humans. *Heredity, 46,* 227–238.

Heath, A. C. (1987). The analysis of marital interaction in cross-sectional twin data. *Acta Geneticae Medicae et Gemellologiae, 36,* 41–49.

Heath, A. C., Kendler, K. S., Eaves, L. J., & Markell, D. (1985). The resolution of cultural and biological inheritance: Informativeness of different relationships. *Behavior Genetics, 15,* 439–465.

Lake, R. I., Eaves, L. J., Maes, H. H., Heath, A. C., & Martin, N. G. (2000). Further evidence against the environmental transmission of individual differences in neuroticism from a collaborative study of 45,850 twins and relatives on two continents. *Behavior Genetics, 30*, 223–233.

Maes, H. H., Neale, M. C., & Eaves, L. J. (1997). Genetic and environmental factors in relative body weight and human adiposity. *Behavior Genetics, 27*, 325–351.

Meyer, J. M., Rutter, M., Silberg, J. L., Maes, H. H., Simonoff, E., Shillady, L. L., Pickles, A., Hewitt, J. K., & Eaves, L. J. (2000). Familial aggregation for conduct disorder symptomatology: The role of genes, marital discord, and family adaptability. *Psychological Medicine, 30*, 759–774.

Nance, W. E., & Corey, L. A. (1976). Genetic models for the analysis of data from the families of identical twins. *Genetics, 83,* 811–826.

Neale, M. C., & Cardon, L. R. (1992). *Methodology for genetic studies of twins and families*. Dordrecht, the Netherlands: Kluwer Academic.

Rutter, M., Pickles, A., Murray, R., & Eaves, L. (2001). Testing hypotheses on specific environmental causal effects on behavior. *Psychological Bulletin, 127*, 291–324.

Silberg, J. L., & Eaves, L. J. (2004). Analysing the contribution of genes and parent-child interaction to childhood behavioral and emotional problems: A model for the children of twins. *Psychological Medicine, 34,* 347–356.

Truett, K. R., Eaves, L. J., Walters, E. E., Heath, A. C., Hewitt, J. K., Meyer, J. M., Silberg, J. L., Neale, M. C., Martin, N. G., & Kendler, K. S. (1994). A model system for the analysis of family resemblance in extended kinships of twins. *Behavior Genetics, 24*, 35–49.