Response to 5% Carbon Dioxide in Children and Adolescents

Relationship to Panic Disorder in Parents and Anxiety Disorders in Subjects

Daniel S. Pine, MD; Rachel G. Klein, PhD; Roxann Roberson-Nay, PhD; Salvatore Mannuzza, PhD; John L. Moulton III, PhD; Girma Woldehawariat, PhD; Mary Guardino

Background: Carbon dioxide (CO₂) sensitivity is postulated to be a familial risk marker of panic disorder (PD). Exaggerated responses to CO₂ inhalation have been reported in adults with PD and their unaffected adult relatives, as well as in clinic-referred children with anxiety disorders.

Objective: To test in a family-based design whether CO₂ hypersensitivity is a familial risk marker for PD and associated with current anxiety disorders in children and adolescents.

Setting and Participants: One hundred forty-two offspring (aged 9-19 years) of parents with PD, major depressive disorder, or no disorder. Forty-five (32%) had a current anxiety disorder, excluding specific phobia.

Design and Main Outcome Measures: Parents and offspring received diagnostic assessments. Offspring underwent 5% CO₂ inhalation at home. Panic symptoms and panic attacks were rated with the Acute Panic Inventory at baseline, while anticipating CO₂ delivery (“threat”), and during CO₂ inhalation. Respiratory rate and volume were measured with spirometry.

Results: No group differences were found in Acute Panic Inventory ratings at baseline or in respiratory measures during threat. Risk for PD was not associated with CO₂ sensitivity (panic symptoms and respiratory physiologic response). During CO₂ inhalation, offspring with anxiety disorders, relative to offspring without anxiety disorders, experienced significantly more panic symptoms and panic attacks, as well as elevated respiratory rates. During threat, panic symptoms were significantly and independently associated with both parental PD and offspring anxiety disorders.

Conclusions: No support was obtained for CO₂ hypersensitivity as a familial risk marker for PD in children and adolescents. Links between childhood anxiety disorders and CO₂ sensitivity were replicated. Familial risk for PD in children and adolescents may be associated with vulnerability to anticipatory anxiety.

Arch Gen Psychiatry. 2005;62:73-80

A Link Between Panic Disorder (PD) and Heightened responses to carbon dioxide (CO₂) inhalation is well documented. Carbon dioxide–enriched air is more likely to provoke panic symptoms and panic attacks as well as an elevated respiratory rate in patients with PD, compared with other disorders. Enhanced CO₂–induced anxiety also has been found in psychiatrically healthy, first-degree adult relatives of patients with PD, leading to the hypothesis that CO₂ hypersensitivity is a familial vulnerability marker for PD. Panic disorder usually develops in adulthood. As a result, studies in psychiatrically healthy adult relatives focus on individuals who have passed through the risk period, excluding those who develop the disorder. Therefore, results from such studies may not be fully informative on the significance of premorbid risk markers. In contrast, juvenile offspring of parents with PD are still at risk for PD and thus provide an informative means for assessing premorbid vulnerability. To our knowledge, no study has reported on CO₂ sensitivity in young offspring of parents with PD.

To investigate biological links between anxiety disorders in children and PD in adults, we previously measured CO₂ hypersensitivity in children. We found that children with anxiety disorders showed greater CO₂ sensitivity than psychiatrically healthy children, confirming similar vulnerability in adults with PD. However, children in these reports were all clinical cases, and studies of nonreferred children are lacking. This family-based study examined whether findings obtained among clinic cases apply to affected nonreferred children. Extension of these findings would confirm the relevance of CO₂ sensitivity in early...
anxiety disorders, fostering research on mechanisms in childhood anxiety disorders.

The current study addresses 2 questions: whether CO₂ sensitivity is greater (1) in offspring at high risk for PD compared with offspring at low risk (ie, whether it is a familial risk marker for PD in children) and (2) in offspring with a current anxiety disorder compared with offspring without an anxiety disorder. First, the study tests the hypothesis that the offspring of parents with PD will exhibit during CO₂ exposure elevated panic symptoms, elevated frequency of panic attacks, and elevated respiratory rate. Specifically, we hypothesized that, relative to responses in offspring of psychiatrically healthy parents, CO₂-related responses would be perturbed in offspring of parents with PD but not in offspring of parents with only major depressive disorder (MDD). We included children of parents with MDD to determine whether correlates of parental PD relate to parental PD specifically rather than parental psychopathology in general. Second, we predicted that offspring with ongoing anxiety disorders (except specific phobia) would show CO₂ hypersensitivity compared with offspring without anxiety disorder. We also examined panic symptoms and respiratory physiologic response immediately before CO₂ exposure (anticipation/threat). Based on prior findings, elevated panic symptoms were expected in offspring of parents with PD, as well as in those with an anxiety disorder.

METHODS

SUBJECTS

Subjects consisted of 142 offspring, aged 9 to 19 years, of parents with PD, MDD, and no mental disorder. Exclusion criteria included history of psychosis, mania, pervasive developmental disorder, current use of psychotropic medication, IQ less than 70, medical conditions that could affect CO₂ response, and residence outside the New York City, NY, metropolitan area. Medical record reviews at outpatient clinics identified parents with PD and/or MDD. Comparison parents were identified from a pediatric dental clinic or using acquaintance methods described elsewhere. Approximately half of comparison parents were recruited from each source. Except for diagnostic status, inclusion/exclusion criteria were identical for probands and comparators.

Parents were drawn from cohorts identified in earlier studies. Data were collected as part of a 2-hour session. The current study included assessments of memory and a physical examination, as well as exposure to facial photographs depicting various emotions; the session ended with the CO₂-inhalation procedure. Informed consent/assent was provided by all participants. Subjects were told that the assessment would involve periods of room air and CO₂ breathing that might induce anxiety. This information was relayed at the time of consent, at the initiation of the in-home assessment, and immediately before the procedure. Although conveying this information 3 times might enhance anticipatory anxiety, it is typically done⁵ for families to be fully cognizant of risks of anxiety induction during CO₂ exposure.

From 290 families with 497 children identified from previous studies, 220 children met study criteria, but 78 (35%) declined participation because of reservations about the CO₂ procedure. All 78 declined when first informed about the potential for anxiety induction; none did so subsequently. This report is based on 142 offspring from 93 families. Offspring who participated in CO₂ inhalation did not differ from those who declined participation in terms of age, sex, ethnicity, social class, or parental or child psychiatric diagnosis.

ASSESSMENTS

Diagnostic Assessment of Parents

Parents were administered the Structured Clinical Interview for DSM-III-R by trained psychologists blind to offspring mental status. Parents were interviewed individually when both parents were available. Interviewers wrote clinical narrative summaries documenting the lifetime DSM-IV diagnoses formulated. Fidelity of the interview was monitored through expert review of narratives. Offspring born to at least 1 parent with PD were considered at high risk; low risk was defined as absence of PD in both parents. Comorbidity in familial risk was also based on diagnoses in either parent. Thus, an offspring at risk for both PD and MDD might have 1 parent with both disorders or 1 parent with PD and another with MDD. Of 40 such offspring, 37 were born to 1 parent with both conditions. None of 24 offspring born to parents with only PD had 2 affected parents; 22 of 52 offspring born to parents with only MDD had 2 affected parents.

Diagnostic Assessment of Offspring

Offspring diagnosis relied on parent and self-reports. In the month before CO₂ inhalation, parents and offspring were interviewed in their home. Trained psychologists blind to parent diagnoses administered the Parent As Respondent Informant Schedule to parents and a child version to offspring. Different staff conducted parent and offspring interviews, as in our prior family studies. Fidelity of the interview was monitored through audiocassettes. Clinicians wrote clinical narrative summaries documenting DSM-IV diagnoses; these were blindly reviewed by an expert clinician for accuracy. A review by 2 raters of randomly selected parent and child interviews (50 from each group) yielded acceptable reliability for anxiety and mood disorders (κ >0.65). Diagnoses were considered present if either the parent or the child report justified a diagnosis.

CO₂-INHALATION PROCEDURE

The decision to conduct CO₂ inhalation in the home resulted from experiences in a prior community-based CO₂ study, where many families were willing to participate only in their home. The procedure was supervised by a physician and technician blind to all psychiatric data. Testing occurred in a room selected by the family, using procedures adapted from those described previously. The current study used a portable apparatus developed for an initial study of adult PD. The procedure involved breathing through a face mask for 25 minutes. Room air initially was delivered for 10 minutes, followed by 15 minutes of 3% CO₂. Respiration was monitored throughout the process using spirometry. Subjects were told that CO₂ exposure could begin at any point.

EVALUATION OF CO₂ HYPERSENSITIVITY

As in prior studies, CO₂ hypersensitivity was quantified through subjective reports of panic symptoms rated on the Acute Panic Inventory (API), which has been validated in pediatric anxiety disorders. It inquires about 23 symptoms that subjects rate as absent (0), mild (1), moderate (2), or severe (3). The API generates a total score (possible range, 0-69) that has served as the dependent measure in previous studies. Ratings were obtained.
at 4 points. The first occurred before placement of the face mask, before respiratory monitoring began; the second, after subjects had been fitted with the face mask and had been breathing room air for 10 minutes; the third API rating was obtained after 5 minutes of CO₂ exposure or before the 5 minutes had elapsed if subjects asked that CO₂ inhalation be discontinued; the fourth was after 15 minutes of CO₂ exposure or at any time during the last 10 minutes if subjects requested that CO₂ inhalation be discontinued. Fourteen subjects asked to discontinue the CO₂ procedure during the initial 5 minutes; therefore, they only have data for the third but not the fourth rating. Because it is important to examine CO₂ sensitivity in subjects who discontinue the procedure, analyses of CO₂ sensitivity rely on the third API rating. An additional 10 subjects asked to stop the procedure after 5 minutes. Before the study, the technician was trained to rate panic attacks, based on the API ratings. As in prior studies, panic attacks required an increase in self-rated anxiety and increases of 1 point or more on at least 4 API symptoms.

Measures of respiration used spirometry methods described in Coryell et al. They involved a continuous recording of breath-by-breath values for respiratory rate and tidal volume, as well as their product (ie, minute ventilation). These data were preprocessed for statistical analyses, involving procedures described previously. Specifically, average values were calculated for the entire 10-minute baseline period and for each of the first 10 30-second periods of the first 5 minutes of CO₂ exposure. Analyses rely on 30-second means, but for ease of data perusal, we present means per minute of CO₂ exposure. Risk-group differences were hypothesized for respiratory rate during CO₂ but not room-air breathing. Since few studies of respiratory physiologic response in pediatric anxiety disorders have been conducted and reports in adult PD indicate differences in other measures of respiratory physiologic response, we also report results for means and variability in tidal volume and minute ventilation.

**DATA ANALYSIS**

Symptomatic measures included the API total score and the rate of panic attacks. These variables tested hypotheses on associations between parent or offspring diagnosis and symptomatic response to the procedure. For respiratory variables, we analyzed mean values during room-air and CO₂ breathing for respiratory rate, tidal volume, and minute ventilation, as well as within-subject standard deviations. These variables tested hypotheses on associations between parent or offspring diagnosis and respiratory sensitivity to CO₂. We also examined total time of CO₂ exposure, as recorded during spirometry. Explanatory variables include fixed effects (sex, age, diagnosis in offspring and parents) and random effects (family and sex). Because the API total score distribution was skewed, analyses rely on log-transformed data (Table 1 and Table 2). Repeated measures in the same individual or separate observations on individuals within the same family do not represent independent observations. As some family studies have done, we applied mixed statistical models for binary/categorical (“panic attack”; request to terminate CO₂ procedure) and continuous (API score, respiratory physiologic response, time of CO₂ exposure) data, following procedures in the SAS PROC-MIXED, GEE, and GENMOD modules (SAS Institute, Cary, NC). so that within-sibling correlations were modeled.

For our primary analysis, 2 sets of models were fit. The initial set examined dependent measures (1) as a function of PD in parents, regardless of offspring diagnosis, and (2) as a function of ongoing anxiety disorders in offspring, regardless of parental diagnosis. Our previous studies had not included children with specific phobia as “affected,” because specific phobias are very common and typically not predictive of PD. Hence, to maintain consistency with previous studies, offspring with only specific phobia but no other anxiety disorder were considered unaffected. A second set of multivariate analyses included both offspring as well as parental diagnosis as predictors of the dependent measure.

Three sets of supplementary analyses followed these primary analyses. First, the purpose of this study was to investigate correlates of risk for PD. Offspring of parents with MDD provided an additional comparison group. Hence, an initial supplemental analysis compared offspring of parents with MDD without PD (n=52) and offspring of psychiatrically healthy parents (n=26), excluding the 64 offspring of parents with PD. None of the contrasts approached statistical significance (P values > .20). Therefore, given the absence of influence of parental MDD on the API score and respiration measures during CO₂ exposure, the high-risk group includes offspring of parents with PD, regardless of parental MDD. Second, prior studies of CO₂ hypersensitivity in adults compare unaffected relatives of patients with PD with unaffected relatives of patients without PD. Thus, to generate comparable data in this study, another set of supplementary analyses stratified subjects based on the presence or absence of an ongoing anxiety disorder in the offspring, prior to comparing dependent measures in offspring with and without parental PD. Finally, we compared API scores and respiratory measures in offspring of PD-plus-MDD vs PD-alone groups. No significant differences were obtained; consequently, these results are not reported.

**RESULTS**

Of 142 offspring, 24 were offspring of parents with only PD; 40, of parents with PD and MDD; 52, of parents with only MDD; and 26, of parents without PD or MDD (psychiatrically healthy parents). Table 1 presents sample characteristics as well as API scores in offspring stratified by parental history of PD and MDD. Groups did not differ in age, sex, IQ, or social class. A total of 45 offspring (32%) met criteria for a current anxiety disorder, excluding specific phobia. This included 19 subjects with only social anxiety disorder, 8 subjects with only separation anxiety disorder, 3 subjects with only generalized anxiety disorder, and 15 subjects with multiple anxiety disorders. Table 1 summarizes data on rates of anxiety disorders in offspring of parents with PD or MDD. To examine the association between parental psychopathology and offspring anxiety diagnosis, a multivariate logistic model treated anxiety disorder in offspring as the dependent variable and parental PD and MDD as predictors. This model found that parental MDD (χ²=6.2; P=.01) but not parental PD (χ²=3.2; P=.07) predicted a significantly increased rate of offspring anxiety disorders, though a trend was found for PD. Parental MDD and PD did not have a significant interaction on offspring anxiety disorders (χ²=1.3; P=.20). Table 1 also presents the frequency distribution of API raw scores and means of the log-transformed API total scores at baseline, before CO₂ exposure (threat), and during CO₂ exposure in offspring risk groups.
Table 2 presents characteristics of offspring stratified by presence of anxiety disorders. Offspring with and without anxiety did not differ significantly on demographic variables (ie, age, social class, or IQ), though there was a nonsignificant excess of girls among offspring with anxiety disorders. Table 2 also presents the API descriptive data in offspring with and without anxiety disorders.

### CO2 EXPOSURE: OVERALL FINDINGS

Of 142 offspring, 132 had sufficient data acquired during CO2 exposure for analysis on all variables. Data for total API score were missing for 1 subject at baseline and 1 subject during CO2 exposure; 10 subjects had brief CO2 exposure, precluding analysis of physiologic data. On the whole, the procedure was not highly anxiogenic, as API scores were not high; across all conditions, only 23 (16%) of 142 offspring had scores of 10 or more.

Table 1 presents API scores during the 3 assessment points: baseline, threat, and CO2 breathing. To verify that the study condition produced significant changes in API symptoms, API scores were compared across baseline, threat, and CO2 exposure across offspring groups. Regardless of risk status or presence of anxiety disorder, offspring exhibited significantly higher API scores during CO2 exposure relative to the other epochs. Table 3 presents means for respiratory rate, tidal volume, and minute ventilation at baseline and the initial 5 minutes of CO2 exposure. All 3 respiratory measures showed robust increases during CO2 exposure (all \(P < .001\)), supporting the internal validity of the procedures through expected physiologic effects.

### OFFSPRING AT HIGH RISK FOR PD: CO2-INDUCED PANIC SYMPTOMS, PANIC ATTACKS, AND RESPIRATORY PHYSIOLOGIC RESPONSE

As noted earlier, symptomatic response to CO2 exposure was examined through self-reports on the API and presence of panic attacks.
### Table 2. Characteristics of Offspring With and Without Ongoing Anxiety Disorders

<table>
<thead>
<tr>
<th>Variable</th>
<th>Anxiety Disorders</th>
<th>Statistic</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample size</td>
<td>45</td>
<td>97</td>
</tr>
<tr>
<td>Age, y, mean ± SD</td>
<td>14.4 ± 3.3</td>
<td>15.2 ± 2.8</td>
</tr>
<tr>
<td>No. (%) female</td>
<td>30 (67)</td>
<td>48 (49)</td>
</tr>
<tr>
<td>IQ, mean ± SD</td>
<td>101 ± 12</td>
<td>103 ± 10</td>
</tr>
<tr>
<td>Social class, mean ± SD*</td>
<td>2.8 ± 1.0</td>
<td>2.7 ± 0.8</td>
</tr>
<tr>
<td>API scores Distribution of scores</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>29</td>
<td>11</td>
</tr>
<tr>
<td>1</td>
<td>7</td>
<td>10</td>
</tr>
<tr>
<td>2</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>3</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>4</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>5</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>6</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>7</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>8</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>9</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>10</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>11</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>12</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>13</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>14</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>&gt;14</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>API score, log-transformed mean ± SD</td>
<td>0.16 ± 0.23</td>
<td>0.41 ± 0.30</td>
</tr>
</tbody>
</table>

Abbreviation: API, Acute Panic Inventory.

*Score on 5-factor Hollingshead scale.
†\(P < .001\) for between-group comparison during threat.
‡\(P < .01\) for between-group comparison during carbon dioxide exposure.

### Table 3. Respiratory Physiologic Response and Anxiety in Offspring and Panic Disorder in Parents

#### Respiratory Physiologic Response Variables

<table>
<thead>
<tr>
<th>Subject Groupings</th>
<th>Baseline, Average for Full 10-min Period</th>
<th>Respiratory Rate, min Following CO₂ Exposure†‡</th>
</tr>
</thead>
<tbody>
<tr>
<td>Offspring anxiety disorder</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes (n = 45)</td>
<td>19.45 ± 0.58</td>
<td>18.43 ± 0.57</td>
</tr>
<tr>
<td>No (n = 87)</td>
<td>18.94 ± 0.45</td>
<td>18.71 ± 0.42</td>
</tr>
<tr>
<td>Parent panic disorder</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes (n = 59)</td>
<td>18.92 ± 0.55</td>
<td>18.26 ± 0.51</td>
</tr>
<tr>
<td>No (n = 73)</td>
<td>19.29 ± 0.52</td>
<td>18.76 ± 0.47</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Tidal Volume, min Following CO₂ Exposure†</th>
<th>Minute Ventilation, min Following CO₂ Exposure‡</th>
</tr>
</thead>
<tbody>
<tr>
<td>Subject Groupings</td>
<td>1</td>
</tr>
<tr>
<td>Offspring anxiety disorder</td>
<td></td>
</tr>
<tr>
<td>Yes (n = 45)</td>
<td>0.68 ± 0.05</td>
</tr>
<tr>
<td>No (n = 87)</td>
<td>0.70 ± 0.06</td>
</tr>
<tr>
<td>Parent panic disorder</td>
<td></td>
</tr>
<tr>
<td>Yes (n = 59)</td>
<td>0.70 ± 0.05</td>
</tr>
<tr>
<td>No (n = 73)</td>
<td>0.68 ± 0.04</td>
</tr>
</tbody>
</table>

Abbreviations: f, respiratory rate; Tv, tidal volume; Ve, minute ventilation.

*Offspring with an anxiety disorder exhibited a significantly \(F_{1,1127}=4.4; P < .05\) elevated respiratory rate during carbon dioxide exposure.
†Effect of time was significant \(P < .001\) for all 3 physiologic response variables.
API Scores
As shown in Table 1, no differences on the API were found at baseline between offspring at high and low risk for PD. No significant differences were obtained during CO2 exposure (F=0.7; P>.20). In contrast, during the anticipatory threat period, high-risk offspring reported significantly higher API scores than low-risk offspring (F=5.9; P<.01).

Panic Attacks
Panic attacks occurred in 16 (11%) of 142 offspring, all during CO2 exposure. The mean±SD API total score among subjects with a panic attack was 12.1±6.5 (range, 5-29) compared with 2.3±3.0 (range, 0 to 15) in those without a panic attack. Frequency of panic attacks did not differ between high-risk (7 [11%] of 64) and low-risk offspring (9 [12%] of 78) (x2=0.20; P>.20).

Respiratory Response to CO2 Exposure
Length of CO2 exposure and rate of request for discontinuation of the CO2-inhalation procedure did not differentiate the high-risk from the low-risk offspring. Thus, 9 (15%) of 59 of the high-risk offspring vs 15 (21%) of 87 of the low-risk offspring asked that the procedure be terminated (x2=1.7; P=.20). Our main hypothesis predicted an elevation in respiratory rate among offspring at risk for PD relative to comparisons during CO2 inhalation. As presented in Table 3, we did not find evidence to support this hypothesis. Risk groups did not differ significantly on respiratory rate (F=3.4; P=.06), tidal volume (F=2.1; P=.14), minute ventilation (F=0.11; P>.20), or ventilatory variability quantified by within-subject standard deviations for respiratory rate (F=0; P>.20), minute ventilation (F=2.1; P=.15), and tidal volume (F=2.6; P=.11).

Offspring with Current Anxiety Disorders: CO2-Induced Panic Symptoms, Panic Attacks, and Respiratory Physiologic Response

API Scores
The API scores at baseline did not differ between offspring with and without anxiety disorders. During CO2 inhalation, total API scores were higher in the anxious vs the nonanxious group, whether the model did (F=5.2; P<.05) or did not (F=5.4; P<.01) control for sex. During the threat epoch immediately before CO2 delivery, offspring with a current anxiety disorder reported significantly greater API scores than those without an anxiety disorder, whether sex was controlled (F=11.2; P<.001) or not (F=10.4; P<.001).

Panic Attacks
Rate of panic attacks was significantly elevated among offspring with anxiety disorders (10 [22%] of 45) compared with anxious offspring without anxiety disorders. The 16 cases consisted of 10 (22%) of 45 with a current anxiety disorder and 6 (6%) of 97 without a current anxiety disorder, a statistically significant difference (x2=4.7; P<.05, controlling for sex).

Respiratory Response To CO2 Exposure
Significantly more offspring with anxiety disorders (14 [31%] of 45) than without anxiety disorders (10 [11%] of 87) asked to terminate the CO2 procedure (x2=4.9; P<.05). Length of CO2 exposure also was significantly lower (F=7.9; P<.01) in offspring with anxiety disorders (mean±SD, 12.4±3.3 minutes) compared with those without anxiety disorders (mean±SD, 14.0±3.4 minutes).

The hypothesis that respiratory rate during CO2 exposure would be increased in children with anxiety disorders was supported (F=4.4; P<.05). In contrast, no significant associations emerged with offspring anxiety diagnosis and respiratory variability, mean tidal volume, and minute ventilation.

Given prior results,16 we also conducted preliminary analyses for specific offspring diagnoses of social phobia, generalized anxiety, and separation anxiety disorders. Abnormal respiratory rate during CO2 exposure occurred only with separation anxiety disorder (F=5.9; P=.01). While associations with separation anxiety disorder also emerged for symptom-based measures of CO2 hypersensitivity, these results should be considered preliminary, given the small sample size (n=14) for cases with separation anxiety disorder, among whom 6 had another comorbid anxiety disorder. We will conduct comprehensive analyses of diagnostic specificity after completing attempts to enrich subgroups with specific disorders to allow more refined analyses.

Offspring Anxiety Disorder and Parental PD
Since API scores during threat were related to both PD in the parent as well as anxiety in the child, a further analysis examined the relationship between API scores during threat and both parental PD as well as offspring anxiety disorder, covarying for offspring sex. A significantly higher level of panic symptoms during threat was independently associated with offspring anxiety disorder (F=10.7; P<.001) and parental PD (F=4.3; P<.05). The association with sex was nonsignificant (F=1.4; P>.20).

No interaction effect on API scores was found between offspring anxiety and parental PD (F=0.3; P>.20), nor between parental PD and parental MDD (F=1.0; P>.20). Similarly, no interactions emerged as predictors of physiologic variables (all P values >.20). Finally, parental PD did not predict symptomatic or physiologic response to CO2 exposure in the subset of cases free of an anxiety disorder. Thus, the presence of parental PD did not differentially influence response to CO2 exposure in offspring with or without an anxiety disorder.

The present study used a family-based design to test whether CO2 hypersensitivity is a familial vulnerability marker for PD. In addition, the current design extended prior findings to nonreferred children and adolescents.
with anxiety disorders to assess whether previous findings of CO₂ sensitivity in referred children also characterized nonclinical cases.

A report of at-risk children and adolescents found faster resting respiratory rate and greater respiratory variability during a baseline period in 14 offspring of parents with PD compared with 10 offspring of unaffected parents. The current study did not find such differences in physiological response at baseline. The absence of a relationship between baseline respiratory physiologic response in children and either ongoing anxiety in the child or PD in the parent argues against the notion that anxiety in young individuals is associated with perturbed respiratory physiologic response under conditions of no or relatively minor stress. Abnormalities in respiratory physiologic response among anxious children and adolescents emerge more consistently under conditions of stress.

Abnormal reactions to CO₂ inhalation have been viewed as indicators of premorbid vulnerability for PD, based on the observation that CO₂ hypersensitivity is found in a range of clinical populations related to PD. For example, it occurs in some pediatric anxiety disorders as well as in psychiatrically healthy, adult, first-degree relatives of patients with PD. We did not obtain support for the hypothesis of CO₂ sensitivity as a familial vulnerability marker. Relative to comparisons, offspring of parents with PD did not report more panic symptoms or display greater respiratory changes during 5% CO₂ breathing. In contrast, evidence for familial vulnerability did emerge during anticipation of CO₂ exposure, immediately prior to CO₂ exposure. Conceptualized as a threatening context, offspring of parents with PD, relative to offspring of parents without PD, exhibited elevated panic symptoms but normal respiratory physiologic response in this situation.

Five studies have examined CO₂ sensitivity in adult offspring of patients with PD. Four used a single breath of 35% CO₂, a concentration more anxiogenic than the 5% CO₂ used in the current study. All found greater panic symptoms with 35% CO₂ exposure in at-risk adult offspring than in comparisons. The fifth study used 35% as well as 5% CO₂; associations between risk for PD and panic symptoms were found with 35% but not 5% CO₂ exposure. Adult relatives of patients with PD in this fifth study also exhibited an abnormal minute ventilation response but a normal respiratory rate response to 5% CO₂ and normal variability in respiratory parameters. Such physiologic findings differ from those typically found in adult PD, where, as in the current study, abnormalities emerge in the respiratory rate but not the minute ventilation response to 5% CO₂. Other abnormalities, such as enhanced variability in respiration or perturbations in minute ventilation, emerge with less consistency.

Inconsistencies in CO₂-inhalation studies of familial vulnerability may relate to several methodological factors. First, 5% CO₂ is a less-potent panicogen than 35% CO₂, as shown by higher rates of panic attacks with 35% vs 5% CO₂ in patients with PD as well as psychiatrically healthy adults. Statistical power was limited in the current study by the low rate of panic attacks, and studies obtaining higher rates of panic attacks would possess greater statistical power. In light of these considerations, enhanced responses to 5% CO₂ might occur only in subjects with ongoing anxiety disorders, as suggested by negative results among offspring with PD in the current study and the lone study of 5% CO₂ in adult relatives of patients with PD. Thus, it may be that CO₂ hypersensitivity, as a family-based marker of vulnerability to PD in at-risk children and adolescents, will be detectable only with higher than 5% CO₂ concentrations.

Second, studies in adults with PD have reported contextual influences on respiration. To our knowledge, this is the first study to examine CO₂ response in the home, a venue that is less threatening than a laboratory. However, contextual factors alone cannot account for the current negative findings, since parent-PD differences emerged during threat, which occurred in the same relatively “safe” context. In addition, we replicated findings of greater CO₂ sensitivity among children and adolescents with anxiety disorders. Thus, consistent with our hypothesis, a significant relationship between CO₂ response and current anxiety disorders was found. Similarly, it is conceivable that the confluence of risk in offspring through the presence of an anxiety disorder and a parental history of PD would enhance the detection of vulnerability. Such was not the case. Finally, during room-air breathing while anticipating CO₂ exposure, ongoing anxiety disorders in the offspring and PD in a parent were independently associated with significantly higher levels of panic symptoms. Offspring anxiety and parental PD had additive rather than interaction effects. While these associations are consistent with findings in pediatric anxiety disorders, they differ insofar as adults with PD have more marked responses to CO₂ than to its anticipation (before CO₂ exposure).

Previous reports compared healthy volunteers to children and adolescents referred to clinics for treatment of anxiety disorders. All found elevated panic symptoms before and during CO₂ inhalation in patients, with larger effect sizes during CO₂ exposure. Across these studies, rate of panic attacks was 1% in 70 psychiatrically healthy subjects and 32% in 69 patients. In the current study, we found a rate of 6% in unaffected subjects and 22% in those with anxiety disorders. In terms of physiologic data, as in at least 7 prior studies of adult PD, studies in children and adolescents find patients with anxiety to exhibit elevated respiratory rates during CO₂ inhalation.

Current findings should be considered in light of study limitations. Statistical power was limited by the low rate of panic attacks, as well as the relatively small number of at-risk offspring who are likely to develop PD. Some procedures may have increased levels of anticipatory anxiety. For example, to facilitate full disclosure, subjects were informed 3 times about the potential anxiogenic effect of CO₂ inhalation. While this procedure did not influence compliance, it may have increased anticipatory anxiety. Similarly, the CO₂-inhalation procedure was completed following a 2-hour assessment that involved tests of memory, a physical examination, and exposure to photographs of faces depicting various emotions. While anxiety elicited by these pro-
pedures is low, these procedures also may have facilitated anticipatory anxiety. Therefore, findings of greater panic symptoms during threat may reflect the combined effects of anticipation of CO$_2$ exposure coupled with an effect of procedural factors that preceded respiratory monitoring. However, these factors are unlikely to account for negative results. Levels of panic symptoms prior to CO$_2$ administration were higher in anxious offspring and those of patients with PD, suggesting that affected or at-risk groups are particularly susceptible to anticipatory anxiety. High levels of anticipatory anxiety are unlikely to contribute to negative findings, given that previous studies in both children and adults demonstrate that anticipatory anxiety predicts an enhanced, as opposed to a reduced, response to CO$_2$. The current findings are based on at-risk offspring recruited through parents who had sought treatment for PD or MDD. Similar sampling strategies have been used in studies of familial risk, but they are vulnerable to referral biases. Importantly, 35% of eligible subjects declined participation. Though participants did not differ from nonparticipants in terms of parental or offspring psychopathology, it is possible that those who declined may have been particularly vulnerable to CO$_2$ exposure, and their absence may have contributed to type II errors. In sum, this study of children and adolescents obtained support for CO$_2$ sensitivity as a marker of anxiety but not as a familial risk factor for PD.

Submitted for Publication: January 26, 2004; final revision received May 19, 2004; accepted June 10, 2004. 
Correspondence: Daniel S. Pine, MD, National Institute of Mental Health, Bldg 15-K, Room 110, MSC-2670, Bethesda, MD 20817-2670 (daniel.pine@nih.gov).

Funding/Support: This study was supported by grant R01 MH-59171 from the National Institute of Mental Health, Bethesda, Md, and an Independent Investigator Award National from the Alliance for Research on Schizophrenia and Depression, Great Neck, NY (Dr Pine).

REFERENCES

30. van Beek N, Griez E. Reactivity to a 35% CO$_2$ challenge in healthy first-degree relatives of patients with panic disorder. Arch Gen Psychiatry. 2000;57:960-967.