American Journal of Medical Genetics (Neuropsychiatric Genetics) 114:260–268 (2002)

Quantitative Trait Locus for Reading Disability on Chromosome 6p Is Pleiotropic for Attention-Deficit/Hyperactivity Disorder

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Comorbidity is pervasive among both adult and child psychiatric disorders; however, the etiological mechanisms underlying the majority of comorbidities are unknown. This study used genetic linkage analysis to assess the etiology of comorbidity between reading disability (RD) and attention-deficit hyperactivity disorder (ADHD), two common childhood disorders that frequently co-occur. Sibling pairs ($N = 85$) were ascertained initially because at least one individual in each pair exhibited a history of reading difficulties. Univariate linkage analyses in sibling pairs selected for ADHD from within this RD-ascertained sample suggested that a quantitative trait locus (QTL) on chromosome 6p is a susceptibility locus for ADHD. Because this QTL is in the same region as a well-replicated QTL for reading disability, subsequent bivariate analyses were conducted to test if this QTL contributed to comorbidity between the two disorders. Analyses of data from sib pairs selected for reading deficits revealed suggestive bivariate linkage for ADHD and three measures of reading difficulty, indicating that comorbidity between RD and ADHD may be due at least in part to pleiotropic effects of a QTL on chromosome 6p.

KEY WORDS: reading disability; ADMD; QTL; linkage; comorbidity

INTRODUCTION

Reading disability (RD) and attention-deficit hyperactivity disorder (ADHD) are two of the most common childhood disorders, each occurring in approximately 5% of the population [American Psychiatric Association, 1994]. Results from twin studies suggest that both RD and ADHD are significantly attributable to genetic influences. Specifically, estimates of the heritability of individual differences (h^2) are moderate to high for both ADHD symptoms ($h^2 \approx .75$) [e.g., Gjone et al., 1996; Eaves et al.,1997; Levy et al., 1997; Nadder et al., 1998] and reading achievement ($h^2 \approx .50$) [e.g., Brooks et al., 1990; Wadsworth et al., 1999]. Moreover, twin studies of the etiology of extreme scores suggest that group deficits in reading are moderately heritable [e.g., DeFries and Alarcón, 1996; Wadsworth et al., 2000] and that extreme ADHD scores are attributable primarily to genetic influences [Gillis et al., 1992; Stevenson, 1992; Gjone et al., 1996; Levy et al., 1997; Willcutt et al., 2000a].

Comorbidity of RD and ADHD

RD and ADHD co-occur more frequently than expected by chance; 25–40% of children with either RD or ADHD also meet criteria for the other disorder [e.g., August and Garfinkel, 1990; Semrud-Clikeman et al., 1992; Willcutt and Pennington, 2000]. However, the etiology of this association is not well understood.

Grant sponsor: National Institute of Child Health and Human Development (NICHD); Grant numbers: HD-11681, HD-27802, HD-04024; Grant sponsor: National Institute of Mental Health (NIMH); Grant number: MH-38820; Grant sponsor: NIMH National Research Service Award; Grant number: MH-12100.

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Received 16 May 2001; Accepted 24 October 2001
DOI 10.1002/ajmg.10205

RD and ADHD are significantly comorbid in both clinical and community samples [e.g., Fergusson and Horwood, 1992; Willcutt and Pennington, 2000], indicating that it is not a selection artifact. Similarly, because RD is assessed by cognitive tests, whereas ADHD is assessed by behavioral ratings, the relation between RD and ADHD does not appear to be due to shared method variance [e.g., Willcutt et al., 2001b].

Several additional explanations have been proposed to account for comorbidity of RD and ADHD. In a family study of the biological relatives of children with ADHD, Faraone et al. [1993] found that comorbidity between RD and ADHD was best explained by cross-assortative mating for the two traits. Alternatively, the phenocopy hypothesis proposed by Pennington et al. [1993] suggests that RD may lead to the phenotypic manifestation of ADHD in the absence of the etiological influences typically associated with ADHD in isolation. For example, a child might appear to be inattentive or hyperactive in the classroom due to frustration elicited by difficulties with reading, rather than cognitive deficits that are frequently associated with ADHD in the absence of RD.

A final competing hypothesis suggests that comorbidity between RD and ADHD is due to common etiological influences. The influence of genes on behavior is likely to be pleiotropic, such that the same genes affect more than one phenotype [e.g., Falconer and MacKay, 1996]. For example, the gene for albinism in mice is also associated with significantly higher levels of emotionality [e.g., DeFries et al., 1966; Turri et al., 2001]. Consistent with this hypothesis, recent twin studies revealed significant bivariate heritability of RD and ADHD, suggesting that the pleiotropic effects of a common gene or genes increase susceptibility to both disorders [Stevenson et al., 1993; Light et al., 1995; Willcutt et al., 2000b]. However, genes that influence both disorders have not been localized.

A series of recent studies have attempted to identify genes that contribute to reading difficulties. Smith et al. [1991] first reported evidence suggesting that a gene on chromosome 6 might influence reading deficits. Subsequently, Cardon et al. [1994, 1995] used an interval mapping technique to localize a quantitative trait locus (QTL) for RD to chromosome 6p21.3. Although one subsequent study did not find evidence of a QTL in this region [Petryshen et al., 2000], this localization has now been confirmed in three independent samples [Fisher et al., 1999; Gayán et al., 1999; Grigorenko et al., 2000]. Moreover, these subsequent studies revealed significant linkage for both overall reading ability and several specific reading and language skills that influence reading ability. This represents one of the most consistently replicated linkages in genetic studies of complex traits with respect to both phenotype definition and chromosomal refinement [e.g., Flint, 1999]. Therefore, based on the finding that common genetic influences contribute to comorbidity of RD and ADHD, the present study tested if the QTL for RD on chromosome 6p is also associated with increased susceptibility to ADHD.

MATERIALS AND METHODS

Participants

Subjects participated in the Colorado Learning Disabilities Research Center (CLDRC) twin project, an ongoing study of the genetics of learning disabilities and ADHD [DeFries et al., 1997]. The present analyses used a subset of the sample analyzed by Gayán et al. [1999], for whom ratings of ADHD symptoms were also available (90%). There were no significant differences between pairs with and without ADHD ratings on measures of reading, family history of reading problems, gender ratio, age, or socioeconomic status.

Pairs of dizygotic (DZ) twins and any biological siblings of the twins were recruited if at least one twin in the pair exhibited evidence of reading difficulties in their school records. Such evidence included low standardized test scores, referrals for academic tutoring, or special education placement. Because children with learning difficulties are at increased risk for ADHD [e.g., Fergusson and Horwood, 1992; Willcutt and Pennington, 2000], we anticipated that the prevalence of both reading difficulties and ADHD would be higher in this enriched sample than in the general population. The total sample included 63 families of two siblings, 8 families of three siblings, and 2 families of four siblings, yielding a total of 85 independent sibling pairs. The mean age of the participants was 11.4 years (SD = 2.5; range = 8–18), and 44% of the participants were female.

Procedures

The reading measures were administered in an initial testing session conducted at the Department of Psychology and Institute for Behavioral Genetics, University of Colorado, Boulder. The measure of ADHD was obtained during a second session scheduled approximately 2 weeks later at the Department of Psychology, University of Denver. All measures at both sites were administered by trained examiners with at least a bachelor's degree who had previous experience working with children. Examiners who administered the ADHD interview were graduate students in the clinical child psychology doctoral program at the University of Denver. All examiners were unaware of the diagnostic status of the child and the results of testing conducted at the other sites.

Measures of Reading

As part of the overall study, each individual completed an extensive battery of tests of reading and language skills [e.g., Olson et al., 1994; Gayán et al., 1999]. Phenotypic correlations between the ADHD composite score and all reading measures were significant, ranging from $-.26$ to $-.40$. Therefore, to minimize the likelihood of false positives due to multiple testing if all reading phenotypes were included in the present study, three reading phenotypes were selected for analysis. An overall reading composite score was utilized because it best approximates the method that is typically used to define RD in the literature and in

clinical settings. In addition, measures of orthographic coding and phonological decoding were analyzed because these phenotypes exhibited the strongest evidence for univariate linkage in this region in our previous study [Gayán et al., 1999].

The composite reading discriminant function score (DISCRIM) was computed for each individual employing weights obtained from a discriminant function analysis of subtests from the Peabody Individual Achievement Test (PIAT) [Dunn and Markwardt, 1970] in independent samples of nontwins with and without a history of RD [DeFries, 1985]. Orthographic choice, the ability to recognize words' specific orthographic patterns, was assessed by an 80-trial forced choice task that requires the individual to identify a target word vs. a phonologically identical nonword foil (i.e., *rain, rane*) [Olson et al., 1989]. Phonological decoding, the ability to decode unfamiliar printed words, was measured through an 85-item oral nonword reading task [Olson et al., 1994].

Measure of ADHD

Because many subjects participated in the study prior to the development of the criteria for DSM-IV ADHD [American Psychiatric Association, 1994], the DSM-III [American Psychiatric Association, 1980] version of the parent-report Diagnostic Interview for Children and Adolescents (DICA) [Herjanic and Reich, 1982] was used. The DICA consists of dichotomous items that ask parents to indicate whether or not their child exhibits each of the 16 symptoms of DSM-III ADHD. The inter-interview reliability of the DICA is reported to be .82, and diagnoses based on the DICA have been shown to be concordant with blind clinical assessments approximately 90% of the time [Welner et al., 1987].

A principal axis factor analysis in our sample indicated that symptoms of DSM-III ADHD comprise moderately correlated factors of inattention and hyperactive/impulsive symptoms [Willcutt and Pennington, 2000], consistent with the bi-dimensional model described in DSM-IV [e.g., Lahey et al., 1994]. However, because similar results were obtained when ADHD symptoms were subdivided into separate inattention and hyperactivity/impulsivity factors, only results for the total ADHD score are described in this report (results of analyses of inattention and hyperactivity/impulsivity are available from the first author upon request).

DNA Markers

Eight informative DNA markers spanning a region of 14.7 centimorgans (cM) on the short arm of chromosome 6 were used for these analyses (Table I). These markers span the region reported in previous 6p linkage studies of RD [Cardon et al., 1994, 1995; Fisher et al., 1999; Gayán et al., 1999; Grigorenko et al., 2000; Petryshen et al., 2000]. Siblings and both parents from each family were genotyped for these markers from blood/cheek samples using published methods [Idury and Cardon, 1997]. MAPMAKER/SIBS [Kruglyak and Lander, 1995] was used to estimate the proportion of alleles shared identical by descent (IBD) at each marker ($\hat{\pi}$) for single-marker analyses. For multipoint analyses, MAPMAKER/SIBS uses available information at all markers to estimate the proportion of alleles shared IBD at positions between markers ($\hat{\pi}_q$). Multipoint tests for linkage were conducted at 0.5-cM intervals across the region of interest (ROI).

TABLE I. Marker Information

Marker	Location	cM[a]	Number of Alleles	Heterozygosity
D6S461	6p22.2	.0	11	.70
D6S276	6p22.2	1.6	14	.76
D6S105	6p22.1	3.9	14	.81
D6S306	6p22.1	5.0	10	.61
D6S258	6p21.33	5.0	12	.69
D6S439	6p21.31	10.1	13	.68
D6S291	6p21.31	11.6	7	.69
D6S1019	6p21.2	14.7	12	.68

[a]Distance in cM distal from D6S461. Chromosomal locations retrieved from the draft assembly of the human genome database at the University of California, Santa Cruz [International Human Genome Sequencing Consortium, 2001; http://genome.ucsc.edu]. Relative distance among markers, number of alleles, and heterozygosity were computed from our sample.

STATISTICAL ANALYSIS

Identification of Probands

Scores on each measure were age regressed and expressed in SD units relative to the estimated mean of the overall population of children. The mean score for the population was estimated from the large database of twins and siblings available at the CLDRC. Participants with eight or more ADHD symptoms were identified as probands because this is the number of positive symptoms that are required to meet criteria for DSM-III ADD with hyperactivity or DSM-III-R ADHD. This cutoff fell slightly more than 1.5 SD above the mean of the distribution of ADHD symptoms in the overall population (SD = 1.56). Therefore, for the bivariate analyses, participants scoring more than 1.5 SD below the mean on each reading measure were considered probands. The number of independent sib pairs selected by this criterion was 48 for ADHD, 50 for orthographic choice, 58 for phonological decoding, and 69 for the reading DISCRIM score.

Linkage Analysis

When a sample is selected because at least one sibling exhibits an extreme score on a trait, the regression model described by DeFries and Fulker [1985, 1988] can be adapted to provide a versatile and powerful test for linkage [e.g., Fulker et al., 1991; Cardon and Fulker, 1994]. If a QTL close to a chromosomal marker influences the selected trait, the scores of co-sibs of selected probands should regress differentially toward the population mean as a function of the number of marker alleles shared IBD with the proband.

For each of the present analyses, all sibs who scored beyond the 1.5 SD cutoff score were selected as

probands. For those cases in which both siblings from a pair scored beyond the cutoff, the pair was double-entered into the data file (i.e., each member of the pair was entered once as a proband and once as a co-sib). This double-entry procedure is most appropriate when a sample has been ascertained using truncate selection [e.g., McGue, 1992; Lyons et al., 1997]. Because the double entry of concordant pairs artificially inflates the sample size, the standard errors of the regression coefficients were corrected prior to tests of significance [Stevenson et al., 1993; Gayán et al., 1999].

The regression model for the univariate case is

$$C = B_1P + B_2\hat{\pi} + K \quad (1)$$

where C is the expected co-sib score, P is the proband score, $\hat{\pi}$ is the estimated proportion of alleles shared IBD at the marker (or $\hat{\pi}_q$ for chromosomal locations between markers), and K is the regression constant. After adjustment of the standard errors of the regression coefficients to correct for the double entry of concordant sibling pairs, the significance of the B_2 parameter provides a statistical test of the extent to which extreme scores are influenced by a QTL linked to the marker.

A simple generalization allows the univariate model to be applied to bivariate data. Rather than comparing the relative similarity of siblings for the same trait, bivariate analyses test if the relation between the proband score on the selected trait and the co-sib score on a second, unselected trait also varies as a function of $\hat{\pi}$. Therefore, if the QTL for RD is also a susceptibility locus for ADHD, the number of symptoms of ADHD exhibited by co-sibs of RD probands would be expected to increase with allele sharing at the susceptibility locus. The regression model for the bivariate case is

$$C_{ADHD} = B_1P_{RD} + B_2\hat{\pi} + K \quad (2)$$

where C_{ADHD} is the predicted co-sib score on the nonselected measure (ADHD) and B_2 tests for pleiotropic effects of the QTL on the two phenotypes.

Because multipoint linkage analysis takes advantage of information from all markers to estimate the proportion of alleles shared IBD at a location, this method has greater power than single-marker analysis when genotyping errors are minimal and estimates of marker order and intermarker distance are accurate. However, genotyping errors or imprecise estimates of intermarker distance or order can alter substantially estimates of significance and location of the susceptibility locus in multipoint analysis [Lincoln and Lander, 1992; Halpern and Whittemore, 1999; Douglas et al., 2000]. Therefore, in this paper we report results of both single-marker and multipoint analyses.

RESULTS

Initial univariate analyses were conducted to test if the QTL on chromosome 6p21.3 is also a susceptibility locus for ADHD. The sibling correlation for symptoms of ADHD was .37, indicating that ADHD is significantly familial in this sample. Results of single-point analyses revealed significant linkage for ADHD to three markers, with the strongest evidence for linkage at marker *D6S105* (Table II and Fig. 1). Multipoint analyses also yielded significant results, with peak linkage between markers *D6S276* and *D6S105* (Fig. 1).

Due to the fact that this sample was initially selected because at least one sibling in a family exhibited evidence of reading difficulties in their academic record, a second set of analyses was conducted to test conservatively if the significant linkage for ADHD was simply a secondary consequence of reading problems rather than a susceptibility to ADHD per se. Scores on the three reading phenotypes were regressed out of the ADHD scores of probands and co-twins, and linkage analyses were conducted on the residual ADHD scores. Evidence of linkage for ADHD in single-marker analyses was weaker but remained significant for markers *D6S105* ($t = 2.39$, $P = .01$) and *D6S276* ($t = 1.82$, $P = .04$), and a trend toward significance remained for marker *D6S306* ($t = 1.55$, $P = .06$). Similarly, multipoint analyses of the residual ADHD scores revealed significant evidence for linkage when the three reading phenotypes were controlled in this manner ($t = 2.10$, $P = .02$). These results suggest that a QTL in this region is associated with elevations of ADHD symptoms independent of reading difficulties.

To test for pleiotropic effects of the QTL on RD and ADHD, bivariate linkage analyses were conducted for each reading measure based on the regression model in Eq. [2]. For each analysis, probands were selected for a score below the 1.5 SD cutoff on the relevant reading measure, and the co-sib's ADHD score was then regressed onto the proband's reading score and the proportion of alleles shared IBD. Both single-marker (Table III) and multipoint analyses (Fig. 2) revealed significant bivariate linkage for ADHD and each of the three reading phenotypes, although results were strongest for orthographic choice. Similar to the univariate analyses, the strongest evidence for bivariate linkage was obtained near marker *D6S105*.

DISCUSSION

This study tested whether the well-replicated QTL for RD on chromosome 6p is also a susceptibility locus for ADHD and whether comorbidity between RD and ADHD is attributable at least in part to the effects of this QTL. Univariate linkage analyses indicated that a QTL on chromosome 6p increases susceptibility to ADHD. Moreover, this result remained significant

TABLE II. Single-Marker Linkage Analyses of ADHD Scores

Marker	B_2 (SE)	t	*P*
D6S461	0.93 (0.75)	1.24	.11
D6S276	1.32 (0.67)	1.97	.03
D6S105	1.91 (0.60)	3.18	.001
D6S306	1.59 (0.79)	2.02	.03
D6S258	1.21 (0.76)	1.59	.06
D6S439	0.91 (0.71)	1.29	.10
D6S291	0.96 (0.76)	1.26	.11
D6S1019	0.89 (0.67)	1.33	.10

P-values indicate one-tailed significance levels.

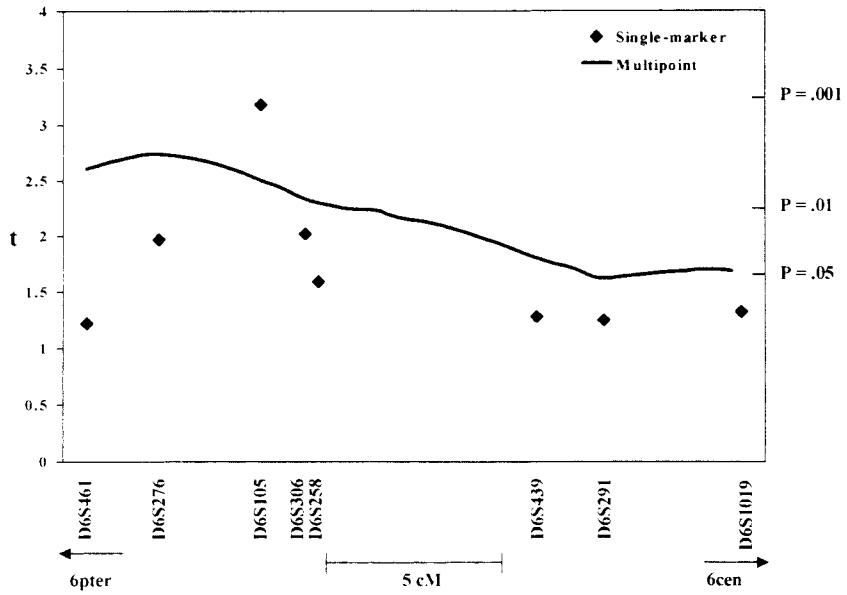

Fig. 1. Single-marker and multipoint linkage of ADHD to eight DNA markers on chromosome 6p ($N = 48$ pairs). P values indicate one-tailed significance levels.

when scores on the three measures of reading were controlled, suggesting that the linkage of ADHD to a QTL in this region is not simply a secondary consequence of reading difficulties. Single-marker analyses revealed peak linkage at marker *D6S105*, and the peak of the multipoint curve fell between markers *D6S276* and *D6S105*.

Bivariate analyses revealed suggestive evidence of bivariate linkage for ADHD and the three reading phenotypes, with the strongest results obtained for orthographic choice. The chromosomal location with the strongest evidence for bivariate linkage was near marker *D6S105* for all three phenotypes. The location of peak linkage for each analysis was within the linkage regions obtained in previous studies of reading difficulties in our sample [Cardon et al., 1994, 1995; Gayán et al., 1999] and others [Fisher et al., 1999; Grigorenko et al., 2000], and all have been mapped outside the classical class I HLA region [MHC Sequencing Consortium, 1999].

The most parsimonious interpretation of these results is that a single QTL in this region has pleiotropic effects that increase risk for both RD and ADHD. This interpretation is consistent with the results of bivariate twin analyses, which suggest that comorbidity between RD and ADHD is attributable primarily to common genetic influences [Stevenson et al., 1993; Light et al., 1995; Willcutt et al., 2000b]. Alternatively, significant bivariate linkage could also be obtained if two or more QTLs with independent effects on RD and ADHD are so close together that they are in linkage disequilibrium. Future fine-mapping and candidate gene analyses will be necessary to conduct a definitive test of these two hypotheses.

Other Explanations for Comorbidity of RD and ADHD

Other researchers have suggested that comorbidity between RD and ADHD could be attributable to

TABLE III. Single-Marker Bivariate Linkage Analyses of ADHD and Reading Scores in Sibling Pairs Selected for Reading Difficulties

	Reading phenotype selected in the proband								
	Reading DISCRIM score			Orthographic choice			Phonological decoding		
Marker	B_2 (SE)	t	P	B_2 (SE)	t	P	B_2 (SE)	t	P
D6S461	0.77 (0.52)	1.48	.07	1.50 (0.65)	2.31	.012	1.31 (0.48)	2.73	.004
D6S276	0.54 (0.43)	1.26	.11	1.38 (0.48)	2.88	.003	0.68 (0.44)	1.55	.06
D6S105	1.18 (0.45)	2.62	.006	1.76 (0.45)	3.91	.0002	1.03 (0.44)	2.34	.012
D6S306	0.96 (0.57)	1.69	.05	1.78 (0.51)	3.49	.0005	0.90 (0.54)	1.67	.05
D6S258	− 0.32 (0.49)	− 0.65	—	1.21 (0.51)	2.37	.011	0.01 (0.48)	0.02	.49
D6S439	0.03 (0.46)	0.07	.47	0.43 (0.52)	0.83	.21	0.42 (0.44)	0.95	.17
D6S291	0.38 (0.51)	0.75	.23	1.25 (0.55)	2.27	.014	0.47 (0.54)	0.87	.19
D6S1019	0.44 (0.47)	0.94	.18	0.56 (0.62)	0.90	.19	0.14 (0.45)	0.31	.38

P-values indicate one-tailed significance levels.

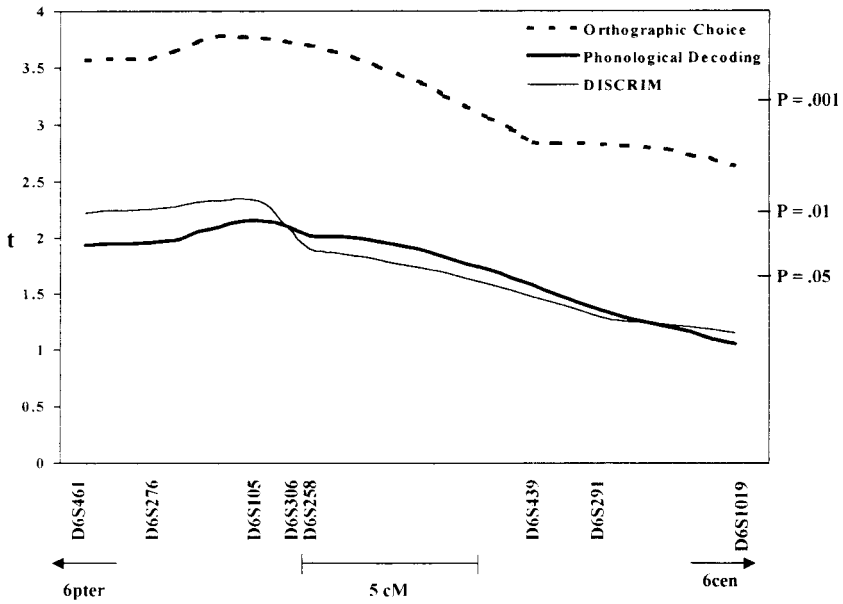

Fig. 2. Bivariate multipoint linkage of reading deficits and ADHD when probands were selected for deficits on orthographic choice ($N = 50$ pairs), phonological decoding ($N = 58$ pairs), and the reading DISCRIM ($N = 69$ pairs). P values indicate one-tailed significance levels.

cross-assortative mating for the two traits [Faraone et al., 1993] or to a causal relation between the two disorders [Pennington et al., 1993]. As noted previously, the cross-assortative mating hypothesis proposes that ADHD and RD are due to distinct etiological influences that are transmitted independently and that comorbidity of RD and ADHD occurs because individuals with ADHD are more likely to reproduce with individuals with RD than would be expected by chance. In contrast, significant bivariate linkage suggests that the same risk allele (or multiple risk alleles in linkage disequilibrium) increases susceptibility for both disorders. Therefore, while phenotypic cross-assortment may cause some cases of comorbidity between RD and ADHD [Faraone et al., 1993], the present results suggest that this comorbidity is also at least partially attributable to the effects of a QTL on chromosome 6p.

In a cognitive study of the etiology of comorbidity between RD and ADHD, Pennington et al. [1993] found that individuals with ADHD exhibited significant deficits on executive function tasks but were not impaired on measures of phonological processing. In contrast, children with RD exhibited phonological processing deficits but were not impaired on measures of executive functioning. The group of children with both RD and ADHD exhibited significant phonological processing deficits but were not different from the comparison group on the executive function tasks. Based on the finding that children with RD and ADHD did not exhibit the executive function deficits characteristic of children with ADHD alone, Pennington et al. [1993] proposed that RD and ADHD co-occur because RD causes the phenotypic manifestation of ADHD in the absence of the etiological influences typically associated with ADHD in isolation. This hypothesis is of particular importance for the present analyses, because if having RD causes children to exhibit a secondary phenocopy of ADHD in the absence of the etiological influences typically associated with ADHD, the pattern of results could mimic the pattern indicative of bivariate linkage.

Two results provide evidence against the phenocopy hypothesis in the present sample. First, linkage for ADHD remained significant when all three reading scores were regressed out of the ADHD scores prior to the analysis, suggesting that linkage for ADHD is not simply a secondary consequence of the reading difficulties exhibited by many children in our sample. In addition, most subsequent studies of the cognitive correlates of RD and ADHD have not supported the phenocopy hypothesis [e.g., Robins, 1992; Reader et al., 1994; Nigg et al., 1998], including data from the overall CLDRC sample [Willcutt et al., 2001b]. Instead, these studies have found that the group with RD and ADHD exhibits both the executive function deficits associated with ADHD and the phonological processing deficits associated with RD.

LIMITATIONS

The present results should be interpreted in light of several limitations. Interpretation of the univariate linkage analyses for ADHD is complicated by the fact that the sample was recruited initially because at least one sibling in each family exhibited evidence of learning difficulties in their school records. Because learning difficulties are significantly associated with ADHD [e.g., Semrud-Clikeman et al., 1992; Willcutt and Pennington, 2000], this method of ascertainment yielded a larger number of probands with ADHD than would be expected in an unselected sample. Although the utilization of a sample enriched for ADHD provided greater statistical power for linkage analyses, this

could limit the applicability of the current findings to unselected populations. For example, it is possible that the etiology of ADHD differs depending on the presence or absence of comorbid RD. However, linkage for ADHD remained significant when all three reading scores were controlled, suggesting that although a portion of the variance attributable to this QTL is a shared risk factor for RD and ADHD, the QTL is also associated significantly with ADHD, independent of reading deficits. Nevertheless, the current findings should be interpreted with caution until they can be replicated in a separate sample selected directly for ADHD.

Because the CLDRC twin project has been ongoing for nearly 20 years, we have maintained a measure of DSM-III ADHD and a version of the PIAT older than the edition that is currently available to allow comparisons to be made across the entire sample. Other etiologically informative studies of RD or ADHD have obtained relatively similar results across different samples and measures [e.g., Biederman et al., 1990; Thapar et al., 1995; DeFries and Alarcón, 1996; Eaves et al., 1997; Levy et al., 1997; Faraone et al., 2000; Willcutt et al., 2001a]. However, because more recent measures are available, the current findings warrant replication with a measure of DSM-IV ADHD and a reading test with a more recent normative sample. The assessment of DSM-IV ADHD symptoms will also facilitate analyses to test whether the effect of the QTL in this region differs as a function of ADHD subtype.

FUTURE DIRECTIONS

Further Localization of the QTL on 6p

Linkage analysis provides an essential first step toward the localization of susceptibility loci for complex traits such as RD or ADHD. However, because recombination occurs relatively infrequently between two closely linked genes across a few generations of a family, the resolution of linkage analysis is often not sufficient for fine mapping of the chromosomal location of a QTL [e.g., Roberts et al., 1999; Lake et al., 2000]. In contrast to family-based linkage analysis, allelic association methods test whether the marker locus and QTL are in linkage disequilibrium in an entire population, such that alleles at the marker locus co-segregate with alleles at the susceptibility locus more frequently than expected by chance [e.g., Cardon and Bell, 2001]. Because linkage disequilibrium rarely extends more than 0.5 cM from a marker [e.g., Abecasis et al., 2001], allelic association methods can be used with more densely spaced DNA markers to refine the estimate of the chromosomal location of the QTL within the region of significant linkage. Therefore, we are currently genotyping additional markers across the region of significant linkage on chromosome 6p and flanking regions of the chromosome.

Common Neurocognitive or Physiological Deficit in RD and ADHD

Although the present results provide further converging evidence that common genetic influences increase risk for RD and ADHD, the physiological mechanisms of these genes are unknown. Previous studies have not revealed a neurocognitive deficit or physiological marker that is consistently a risk factor for both disorders [e.g., Willcutt et al., 2001b]. However, few neurocognitive studies have compared RD and ADHD groups on measures of speeded verbal naming, nonverbal working memory, cognitive processing speed, or variability of reaction time. Alternatively, the effects of a pleiotropic gene on chromosome 6 could lead to two distinct syndromes that are largely distinct at the phenotypic and neurocognitive levels of analysis. For example, the gene for albinism in mice is also associated with significantly higher levels of emotionality [e.g., Turri et al., 2001]. Although no obvious candidate genes for RD or ADHD have been identified in this region to date, future molecular genetic studies in humans and animals will determine the function of additional genes in this region. Based on these findings, plausible candidate genes may be identified that are expressed in the brain or influence other developmental processes that are related to RD and ADHD.

Genetic Heterogeneity

In addition to the QTL on chromosome 6, possible QTLs for RD have been localized to regions on chromosomes 1 [e.g., Rabin et al., 1993], 2 [Fagerheim et al., 1999], 15 [e.g., Smith et al., 1983; Fulker et al., 1991; Grigorenko et al., 1997], and 18 [Fisher et al., 2001], and additional QTLs are likely to be identified in genome-wide scans that are currently being conducted [e.g., Fisher et al., 2001]. Similarly, candidate gene studies have demonstrated a significant association between ADHD and polymorphisms in at least five genes in the dopamine system [e.g., Cook et al., 1995; Daly et al., 1999; Eisenberg et al., 1999; Faraone et al., 1999; Barr et al., 2000a], although these results have not replicated in all samples [e.g., Asherson et al., 1998; Castellanos et al., 1998; Barr et al., 2000b]. These results underscore the complexity of the genetic etiology of RD and ADHD and suggest that additional genes are also likely to contribute to comorbidity of these disorders. Therefore, we are presently analyzing these additional chromosomal regions and candidate genes. In future analyses we will test which of these genes contribute independently to RD, ADHD, or their comorbidity and whether epistatic interactions among any of the loci play a significant role in the etiology of these disorders and their overlap.

CONCLUSIONS

Comorbidity is the rule, rather than the exception, for both adult and child psychiatric disorders, but very little is known about the causal mechanisms underlying these associations [Caron and Rutter, 1991; Rutter, 1994; Neale and Kendler, 1995]. The present results in a sample of sib pairs in which at least one sib has reading difficulties indicate that the QTL for RD on chromosome 6p is also a susceptibility locus for ADHD and suggest that comorbidity between RD and ADHD

may be due at least in part to pleiotropic effects of this QTL. This result suggests that the boundaries between putatively distinct diagnoses may be blurry, with the same genetic influences conferring risk for more than one disorder. In some cases, such findings may indicate that two disorders may be better conceptualized as alternate forms of the same disorder, whereas in other cases common risk factors may contribute to two or more distinct disorders. In either case, methods such as those described in this paper provide an important tool that can be used in future studies to improve the validity of the diagnostic nosology of psychiatric disorders by revealing the etiology of comorbidity between complex syndromes.

ACKNOWLEDGMENTS

Supported in part by program project and center grants from the National Institute of Child Health and Human Development (NICHD; HD-11681, and HD-27802) to J.C.D., grants from the National Institute of Mental Health (NIMH; MH-38820) and NICHD (HD-04024) to B.F.P., and NIMH National Research Service Award to E.G.W (MH-12100). We thank the school personnel and families who participated in the study and Helen E. Datta, Terry Goldhammer, and Deborah Porter for database management.

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