Supplementary Information For: A Genome-scan for Loci Shared by Autism Spectrum Disorder and Language Impairment

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Supplementary Methodological Information

Families

Criteria for family recruitment for the study included: 1) at least one individual with a diagnosis of autistic disorder with no known genetic cause (i.e. Fragile X or Rett's), 2) at least one other individual who met criteria for *specific language impairment* (SLI), 3) at least three other family members willing to participate, and 4) English as the primary language of all individuals participating. Prior to behavioral testing, all subjects gave informed consent conforming to the guidelines for treatment of human subjects at Rutgers University. All subjects were tested with a comprehensive neuropsychology battery administered by an experienced psychometrician, speech language pathologist, or psychologist.

Autism proband criteria: To be identified as the Autism Proband, the etiology must be unknown (for example, no Fragile X or Rett's) and the individual was required to meet the cut-off for Autism or Autistic Disorder on at least two of the three following measures (all were administered to all autism probands):

1) Autism Diagnostic Interview-Revised, 2) Autism Diagnostic Observation Scale,

3) Diagnostic and Statistical Manual-IV.

SLI proband criteria: In order to be identified as an SLI proband, a person had to meet the following inclusionary/exclusionary criteria:

1) A core standard score of <= 85 on the age appropriate version of the Comprehensive Test of Language Fundamentals [1,2]; or subtest scores of at least one standard deviation below peers on 60% of all language measures plus a significant history of language and reading difficulties as measured by at least 2+ years of intervention and/or previous childhood diagnosis of language and/or reading impairment.

2) A non-verbal IQ >= 80 on the Wechsler Abbreviated Scale for Intelligence [3] and always greater than the Core Language Score of the Comprehensive Test of Language Fundamentals.

3) Hearing within normal limits [positive identification of 500 Hz at 30 dB (SPL), and 1000, 2000, and 4000 Hz at 20 dB (SPL)].

4) No motor impairments or oral structural deviations affecting speech or non-speech movement of the articulators as assessed by a speech-language pathologist.

5) No history of autism or frank neurological disorders such as mental retardation, seizure disorder, or brain injury as determined from parental report. When autism spectrum behaviors were suspected upon parental interview or if observed by the Speech Language Pathologist during the language assessment, the ADI-R and ADOS were administered to formally rule out ASD.

6) Native English speaker with English as the primary language spoken at home.

Measures

All family members as well as higher functioning family members with ASD received age appropriate measures of language and reading. See Table S2 for a summary of test and subtests. Briefly, the standardized language and reading measures in the battery included:

a. The Clinical Evaluation of Language Fundamentals (CELF-4 and CELF Preschool)[1,2],- A Core Language Score is derived from 3- 4subtests scaled scores (age depending) that address areas of language comprehension, expression, and structure.

b. The Comprehensive Assessment of Spoken Language (CASL)[4],- contains subtests addressing metalinguistic language skills that tap into complex language include abstraction, inference, and also include a subtest that addresses the pragmatic aspects of language. *These areas are of great relevance to older children and adults as well as higher*

functioning individuals with autism who may be challenged by meaning that cannot be accessed directly through lexical and grammatical information; these areas of language are not assessed by most other standardized language measures.

c. The Comprehensive Test of Phonological Processing (CTOPP)[5], Elison and Non-word Repetition subtests only. *The Elision subtest was used to measure deletion and phonological manipulation of sounds in words while the Non-word Repetition task measured phonological short-term memory; both have a strong documented relationship with oral language abilities and reading.*

d. Gray Oral Reading Tests (GORT-IV)[6], assesses oral reading rate, accuracy, and comprehension.

e. The Woodcock Reading Mastery Tests-Revised[7]- Word Attack and Word Identification subtests only, *Subjects age 6 and older received the Word Attack (non word reading) subtest consisting of mono- and polysyllabic pseudowords to assess decoding abilities, and the Word Identification subtests, single word reading of real words arranged in order of increasing difficulty.*

f. The Wide Range Achievement Test 3 (WRAT) [8]- Spelling subtest only.

Genotyping

DNA samples were obtained in most cases from cell lines established from peripheral blood by the Rutgers University Cell and DNA Repository (RUCDR) as part enrolling subjects into the National Institute of Mental Health Autism Collection. For a few subjects who did not consent to drawing blood or for whom cell lines were not successfully established (N=5), DNA was extracted from saliva using Oragene DNA sample collection kits using the recommended protocol in our lab.

Samples were genotyped using Affymetrix Axiom™ 1.0 arrays by the RUCDR. Genotype calling was conducted on 567,893 SNP genotypes on 440 individuals with the Affymetrix Power Tools software package using the Axiom™ GT1 algorithm, which incorporates a novel modification of the BLRMM-P algorithm. Quality control on SNP genotypes was conducted as described previously [9]. Briefly, SNPs and individuals with high missing rates were first excluded from further analysis (missingness >= 0.1). Call rates for saliva DNA samples were slightly lower than compared to cell line derived DNA (98.2% versus 99.4%) but no systematic differences were observed in copy number, sex determination, Mendelian inconsistency rate or other indicators of quality. The linkage markers (also used for ancestry clustering) were chosen from the subset of markers, post quality control, that had minor allele frequency > 0.3, were on average 0.3 cM apart, and had negligible linkage disequilibrium as measured by $r^2 < 0.2$. Marker allele frequencies were estimated using the maximum likelihood option in MERLIN v 1.11[10]. Marker-to-markers linkage disequilibrium was estimated using PLINK v1.07[11] using unrelated persons (founders) in the dataset. These SNPs were used in a relationship checking analysis by RELCHECK [12]. After correcting inconsistencies in the sample IDs or analysis files, all SNPs were tested for Hardy-Weinberg equilibrium (and dropped if P<.001) and Mendelian inconsistencies (dropped if rate > 0.05). Samples with missingness > 0.03 were dropped from further analysis. SNPs with missingness > 0.05 were dropped from further analysis. Two sets of duplicate samples yielded genotype concordance of 99.82% on SNPs from the final quality controlled set. Additionally, 36 samples were also genotyped with an Affymetrix 250k array with 99.76% concordance for 35,094 SNP in common to both arrays. For association analysis, SNPs with minor allele frequency < 0.05 were not included in the analysis.

Statistical Analysis

For all language measures, standardized scores provided with the test were used in downstream analysis. The Social Responsiveness Scale has two forms, a 65-item child form that is standardized using T-Scores and an adult version that has been minimally modified from the child version to make the wording more appropriate but has not been standardized to a T-Score. We therefore used total raw scores assuming both the adult and child data reflect the same underlying quantitative trait. The cut-off for a dichotomous trait social deficit (SRS-DT) was 54 for males and 45 for females which is equivalent to the T-score > 60 criteria used to identify mild impairment in children. Analysis using a graded response model indicated this procedure did not lose appreciable sensitivity compared to item response theory based scoring (Spearman's rho = 0.99). The Y-BOCS had three versions of the test by age and diagnostic status (adult, child and PDD). The PDD scale has only half the number of items as the other scales. We used percent of the maximum possible score for each scale as a single quantitative trait.

Derivation of phenotypic factor scores for quantitative trait analysis. Factor analysis of 21 phenotypic measures stratified across age bands (N=19 for ages 5-8; N=20 for ages 9-13; N=19 for ages 13+) as some language measures are only given at certain ages, though N=17 are administered to all subjects. We used the matrix of genetic correlations from SOLAR v4.3 [13] as the basis for a genetic factor analysis. Since the matrix was not itself positive definite, we first determined the closest symmetric positive definite matrix, utilizing Higham's method [14], and worked with that matrix. We used parallel analysis to settle on 3 factors [15]. With no *a priori* reason to think that the factors would be orthogonal, we used the oblimin oblique factor rotation. We then determined factor scores for each individual; for individuals with missing data, we created 32 imputations,

calculated factor scores based on those imputations, and used the mean score. For individuals reliant on imputations we set all factor scores to missing if any factor had a standard deviation greater than 0.25 standard units across imputations for that individual.

Ancestry checking. Population ancestry was examined through principal components analysis as implemented in EIGENSTRAT, which computes principal components analysis scores over the input SNP data, 8068 SNPs from the linkage map as defined above. All samples that passed quality control (N=440) and all available HapMap samples with at least 85% of the 8068 linkage analysis SNPs were analyzed to provide clear references for the 3 major continental groupings. The first 4 principal components were visualized graphically with HapMap samples color-coded. Outliers were defined as prescribed in the EIGENSTRAT documentation [16].

Linkage/association analysis methods. We chose the PPL framework to conduct all analyses. Historically the PPL was developed as a linkage analysis method (indeed PPL stood for posterior probability of linkage), which was an improvement upon traditional categorical trait LOD score analysis using the pedigree likelihood but could account for multiple trait models (additive, dominant, recessive and any single locus variation between) without inflationary effects of multiple testing or parameter maximization. This was accomplished by using Bayesian statistical tools whereby trait parameters were removed from the likelihood by integration (not maximization as is done in commonly used statistical frameworks) as nuisance parameters. However, the same underlying likelihood need only be slightly modified for association analysis, even on non-pedigrees such as case-control datasets. Joint linkage-association is also possible as well as many extension that allow for quantitative traits, imprinting, sex specific recombination rates and epistasis. Rather than use a new acronym for each variation of the initial PPL likelihoods, it is

preferable to use PPL not as an acronym for posterior probability of linkage but as an identifier for a statistical framework that allows for flexible model of complex disease datasets.

Briefly, the PPL is on the probability scale and regardless of the number of parameters in the model the same scale is always in effect. Thus, analysis from any of the PPL variations can all be directly compared to one another and directly interpreted to quantify the relative strength of the evidence for loci from categorical and quantitative analysis [17]. Categorical traits were analyzed as described previously [9]. For quantitative phenotypes we used a PPL threshold model originally designed for the present dataset and empirically evaluated [18,19]. As some persons with autism are non-verbal, have behavioral issues that interfere with quantitative language/reading assessment or perform too poorly for those assessments to be valid, analysis of quantitative data in a traditional framework would require either ignoring those subjects in the analysis (missing data) or setting their quantitative value to a low, but statistically plausible value (arbitrary constant). Instead of ad hoc data imputation, the PPL includes a threshold parameter that assumes the untestable subjects performed below a threshold that is left unspecified in the PPL analysis. The advantage of this method is that we retain power by including the individuals with ASD who were without quantitative data in the analysis. It is always possible to simply remove those individuals with ASD from the analysis as a contrast condition for elucidating the role of ASD at a locus that may be ostensible linked to a language phenotype.

Primary linkage analysis was conducted on each of the three tiers separately and the linkage evidence was sequentially updated across the three tiers to provide a single metric for linkage evidence. While the primary linkage outcome is for Tier I, which contains ASD

and SLI (or in five cases an "autism language impaired" subject), we also examined results from the other two tiers and the results sequentially updated linkage results over all three tiers (similar to a meta-analysis). Association analysis was conducted similarly; however, a family that contained persons that were not of European ancestry (N=1 in Tier III) was dropped from the association analysis, and several trios (one ASD and two parents) with data from our phenotypic battery were included (N=9, added to Tier III). Due to the sample sizes, the primary analysis is the sequentially updated evidence across all three tiers though Tier I is of interest alone.

To determine the extent of SNP haplotype tag coverage under regions of interest define by linkage peaks, all HapMap [20] SNPs segregating in the CEU HapMap population with minor allele frequency >=0.03 within each linkage regions were acquired via bulk download from HapMap Data Release 28 Phase II & III (www.hapmap.org) on NCBI assembly B36 and the dbSNP 126 dataset. Tag SNP panels were generated using the Tagger algorithm as implemented in the client version of Haploview 4.2 [21]. Comparisons were made between the number of additional SNPs that were required to cover each region when the original Axiom™ SNPs were specified as "force includes" (i.e., mandatory tags) and the number of SNPs that were required to cover the same region without specifying any force includes. Supplementary Table 3 summarizes the results. To determine the fraction of each chromosomal region that was successfully tagged when only the original Axiom™ SNPs are considered, we divided the number of Axiom™ SNPs by the total number of SNPs needed to tag the region (Axiom™ SNPs plus additional HapMap SNPs).

Assessing the relative contribution of the three proband types to the final PPL. The maximized LOD (MOD) was calculated for each cM position by subsets for families with the presence of at least one autism nonverbal subject forming the nonverbal family group, from

the remaining families the presence of at least one autism language impaired forming the language impaired family group, with other families classified as the language normal family group. For both linked and unlinked regions, each subset contributes to the MOD score as an increasing function of sample size (the exact function depends on the pedigree structure), but in linked regions the linked subsets make far larger contributions to the MOD score than the unlinked subsets. Therefore, for each genomic position we define:

$$\omega = \sum_i \left| \alpha_{N,i} - \alpha_{MOD,i} \right|$$

where *i* is the subset index, $\alpha_{N,i}$ is the proportion of families in subset *i*, and $\alpha_{MOD,i}$ is the proportion of the total MOD score attributable to subset *i*. The average of ω in unlinked regions, $\omega_{non-peak}$, can be compared to ω_{peak}, calculated for linked regions, where the ratio would be close to 1 if the linked region were homogeneously linked across the subsets and deviate from 1 as any subset contributed more to the final MOD. We examined $\omega_{peak}/\omega_{non-peak}$ using our defined linkage regions versus the remaining chromosomal positions for the language-related linkage peaks on chromosome 15 and 16. The ratios were 1.02 and 1.06 for the two peaks, both consistent with all three subgroups providing the same proportional linkage signal.

Assessing the within-family relative contributions of language impairment and ASD to the final PPL. For language related linkage peaks, we sought to understand the relative contribution of language impairment versus ASD to the observed score by either removing all SLI probands from the analysis or, removing all autism probands from the analysis, by setting those phenotypes to be missing data in the analysis. However, lack of power under these missing data scenarios may confound interpretation. To assess the probability of observing reductions in the PPL as great or greater than those reported, we performed a

permutation study in the linked regions, randomly removing one phenotyped person (unaffected, LI/RI or ASD) from each pedigree and repeating the analysis. The resulting null distribution allows us to test the hypothesis that SLI and/or autism proband status in particular is important for the observed linkage finding. On chromosome 15, only 1% of the permutations resulted in a lower PPL than removing either SLI probands only or autism probands only (which gave roughly equal drops), indicating a significant relationship between SLI, autism and the maximum PPL without power loss as a confounding factor in interpretation. On chromosome 16, the opposite trend was observed, whereby 75% of the permutations resulted in a lower PPL than that obtained by dropping one RI (non-ASD) subject or the autism proband. Therefore, on chromosome 16 there is no evidence to distinguish the drop in the PPL when RI or autism subjects are removed from the drop in the PPL, (presumably) caused by reduced sample size, when subjects are removed randomly without regard to LI or ASD status. Another way to show that SLI and ASD do not provide unique contributions to a linkage peak is to repeat the permutation procedure, but restrict it to affected persons only (i.e., randomly remove one person with either LI (chr 15)/RI (chr 16) or ASD in each pedigree). A non-significant result from the permutation procedure that is conditional on "affected" for removal indicated any affected phenotype is exchangeable in terms of induced power loss. On chromosome 15, the permutation p-values were 0.48 and 0.55 for ASD and SLI respectively, indicating full exchangeability of the two phenotypes for inducing power loss at that locus. On chromosome 16, the results are not as clear. For SLI P=0.42 while for ASD P=0.005, indicating a greater effect of ASD on the linkage signal than SLI at that locus. However, the unconditional permutation failed to indicate an effect of diagnostic status.

Additional work to find the variants that underlie the linkage peak will be necessary to understand the relationship between those variants to SLI and ASD.

Table S1. Demographic Table for All Tiers - Nuclear and Extended Family Profiles

	N	Mean age (SD)	Range
Family members meeting criteria for Autism	77	13.7 (9.6)	4.0-40.0
Family members meeting criteria for ASD	15	8.0 (4.5)	3.1-18.0
Family members meeting criteria for LI only	43	19.8 (20.3)	4.1-79.1
Family members meeting criteria for RI only	8	34.2 (23.6)	6.1-74.0
Family members meeting criteria for LI & RI only	9	30.4 (17.1)	7.1-51.1
All other family members	202	34.6 (16.9)	3.0-80.1
Total	354		

Table S2. Quantitative Cognitive Phenotypic Battery

Language & Cognitive Battery	Function	Age
Clinical Evaluation of Language Fundamentals- (CELF-4)- Core Score		
Concepts and Following Directions	Follow oral directions of increasing complexity	5+
Word Structure	Complete a sentence with the correct morphological inflection	5-8
Recalling Sentences	Imitation of sentences of increasing length and complexity	5+
Formulated Sentences	Create a meaningful sentence using a target word or phrase	5+
Word Classes	Identify two related words and explain	9+
Word Definitions	Define words with appropriate detail	13+
Clinical Evaluation of language Fundamentals-Preschool		
Sentence Structure	Interpret sentences of increasing length and complexity	3-5
Word Structure	Pronoun usage and complete sentences with the correct morphological inflection	3-5
Expressive Vocabulary	Label people, places, and actions	3-5
Comprehensive Assessment of Spoken Language (CASL)		
Non-literal language	Understand indirect and figurative language and sarcasm	7+
Meaning from Context	Infer meaning from linguistic context	11+
Inference	Use real world knowledge infer meaning	7-17
Ambiguous Sentences	Recognize ambiguity and verbalize it	11+
Pragmatic Judgment	Knowledge and use of pragmatic rules	3+
Comprehensive Test of Phonological Processing (CTOPP)		
Elision	Delete syllables and sounds from words to create new words	5+
Non-Word Repetition	Repeat nonsense words of increasing length	5+
Gray Oral Reading Tests (GORT-4)		
Rate	Timed oral reading	6+
Accuracy	Number or errors while reading	6+
Fluency	Combined score of rate and accuracy	6+
Comprehension	Answer questions about what was read	6+
Woodcock Reading Mastery Tests-Revised		
Word identification	Single word sight reading	6+
Word Attack	Nonsense word reading	6+
Wide Range Achievement Test 3 (WRAT3)		
Spelling	Spelling of dictated words	6+
Wechsler Abbreviated Test of Intelligence (WASI)		
Performance IQ	Composite non-verbal subtest scores: Block Design and Matrices	6+

Table S3. Phenotypic correlation matrix

	CELF				CTOPP		GORT					CSL				WRAT	WRM	
	FS	RS	WCE	WCR	EL	NR	AS	CS	FS	RS	ORQ	AS	MC	NL	PJ	Spell	WA	WID
CLFFS	1	0.65	0.58	0.66	0.44	0.16	0.58	0.51	0.55	0.47	0.59	0.55	0.66	0.57	0.65	0.48	0.44	0.54
CLFRS	0.65	1	0.61	0.68	0.51	0.34	0.6	0.6	0.61	0.59	0.68	0.67	0.71	0.64	0.66	0.55	0.48	0.57
CLFWCE	0.58	0.61	1	0.7	0.38	0.2	0.49	0.53	0.49	0.44	0.57	0.57	0.62	0.61	0.6	0.37	0.44	0.49
CLFWCR	0.66	0.68	0.7	1	0.42	0.19	0.63	0.6	0.63	0.62	0.69	0.58	0.71	0.58	0.62	0.53	0.52	0.63
CTOPEL	0.44	0.51	0.38	0.42	1	0.34	0.47	0.52	0.44	0.43	0.5	0.48	0.44	0.42	0.39	0.47	0.5	0.55
CTOPNR	0.16	0.34	0.2	0.19	0.34	1	0.11	0.22	0.16	0.24	0.21	0.29	0.26	0.21	0.23	0.29	0.24	0.27
GRT4AS	0.58	0.6	0.49	0.63	0.47	0.11	1	0.56	0.96	0.82	0.88	0.53	0.54	0.37	0.45	0.66	0.62	0.7
GRT4CS	0.51	0.6	0.53	0.6	0.52	0.22	0.56	1	0.57	0.57	0.82	0.51	0.62	0.52	0.46	0.47	0.46	0.58
GRT4FS	0.55	0.61	0.49	0.63	0.44	0.16	0.96	0.57	1	0.92	0.92	0.55	0.57	0.41	0.49	0.71	0.62	0.71
GRT4RS	0.47	0.59	0.44	0.62	0.43	0.24	0.82	0.57	0.92	1	0.86	0.53	0.56	0.42	0.47	0.73	0.62	0.73
GRT4ORQ	0.59	0.68	0.57	0.69	0.5	0.21	0.88	0.82	0.92	0.86	1	0.61	0.65	0.5	0.55	0.68	0.58	0.71
CSLAS	0.55	0.67	0.57	0.58	0.48	0.29	0.53	0.51	0.55	0.53	0.61	1	0.69	0.65	0.6	0.5	0.48	0.56
CSLMC	0.66	0.71	0.62	0.71	0.44	0.26	0.54	0.62	0.57	0.56	0.65	0.69	1	0.79	0.71	0.49	0.45	0.58
CSLNL	0.57	0.64	0.61	0.58	0.42	0.21	0.37	0.52	0.41	0.42	0.5	0.65	0.79	1	0.75	0.42	0.35	0.49
CSLPJ	0.65	0.66	0.6	0.62	0.39	0.23	0.45	0.46	0.49	0.47	0.55	0.6	0.71	0.75	1	0.38	0.33	0.45
WRAT	0.45	0.65	0.66	0.6	0.62	0.39	0.23	0.45	0.46	0.49	0.47	0.55	0.6	0.71	0.75	1	0.38	0.33
WRMWA	0.35	0.48	0.55	0.37	0.53	0.47	0.29	0.66	0.47	0.71	0.73	0.68	0.5	0.49	0.42	0.38	1	0.59
WRMWID	0.41	0.44	0.48	0.44	0.52	0.5	0.24	0.62	0.46	0.62	0.62	0.58	0.48	0.45	0.35	0.33	0.59	1

Table S4. Factor loading for the derived quantitative traits

Test	Subscale	Factor 1	Factor 2	Factor 3
CELF	Formulating Sentences	0.995	0.132	-0.219
	Repeating Sentences	0.647	0.379	0.315
	Word Classes - Expressive	0.73	0.451	0.011
	Word Classes - Receptive	0.944	0.014	0.106
CTOPP	Elision	0.082	-0.056	0.974
	Nonword Repetition	0.005	0.137	0.9
GORT	Accuracy	0.903	-0.094	-0.015
	Comprehension	0.372	0.653	0.251
	Fluency	0.892	0.039	-0.257
	Reading Score	0.855	0.171	0.198
CASL	Ambiguous Sentences	0.569	0.382	0.405
	Meaning from Context	0.608	0.404	0.112
	Nonliteral Language	0.284	0.769	-0.053
	Pragmatic Judgment	-0.204	0.775	0.506
WRAT	Spelling	0.95	-0.295	0.103
Woodcock Reading	Word Attack	0.848	-0.544	0.234
	Word ID	0.857	-0.111	0.288

Table S5. Fraction of linkage regions tagged for association analysis

Trait	Chromosome	bp range (kb)	Axiom-HapMap overlap	Additional HapMap SNPs needed	Fraction of region tagged
LI*	15	68361-91690	3596	2499	0.52
RI*	16	16022-26460	1468	962	0.55
SRS-QT	14	98547-106332	1444	1037	0.28
SRS-DT	15	93955-100181	2213	1039	0.53
YBOCS	13	52365-69872	2485	1110	0.63

Table S6. Follow-up Genotyping from GWAS

SNP	Chromosome	bp	cPPLD	Trait
rs3835700*	1	35422400	0.0014	f2
rs2279749	3	4772781	0.0114	f2
rs12490375	3	4773489	0.0026	f2
rs3792495*	3	4774443	0.2	f2
rs3792498	3	4776967	0.0024	f3
rs9831960	3	4777573	0.0028	f1
rs11732255	4	184678911	0.0003	f1
rs12646153*	4	184681169	0.0005	f1
rs7658620	4	184684075	0.0004	f1
rs10011451	4	185495233	0.0015	f1
rs12505290	4	185495461	0.0018	f1
rs6854407	4	185496329	0.0015	f1
rs12514616	5	120374182	0.0011	f2
rs983571	5	120375136	0.0900	f2
rs2973177*	5	120375499	0.0072	f2
rs10973702	9	3818714	0.0012	f1
rs16919730*	9	3819188	0.13	f1
rs7029652	9	3819212	0.0003	f1
rs2987740	9	102914868	0.005	f1
rs1360098	9	102915925	0.0027	f1
rs10760791	9	102918028	0.0142	f1
rs7958047	12	39662871	0.06	f3
rs11179592*	12	39720021	0.0025	f3
rs1797990*	12	39803135	0.1	f3
rs1797988	12	39805150	0.18	f3
rs1626744	12	39805182	0.1	f3
rs764350*	14	49731682	0.0003	f2
rs9930784*	16	20602872	0.0038	f2
rs9930741	16	20602987	0.024	f2
rs12928136	16	20604843	0.0039	f2
rs11643793	16	20607517	0.0115	f2
rs764138	16	20650973	0.04	f2
rs11646042	16	20651537	0.004	f2
rs8044864	16	20653822	0.04	f2
rs9923588	16	25849211	0.18	RI*
rs4511540	16	25850199	0.1	RI*
rs8089400	18	43417118	0.18	LI*
rs8088661*	18	43417377	0.0012	LI*

*These SNPs were on the Axiom array and also had PPLD values > 10%, the remaining SNPs were genotypes as surrogates in high linkage disequilibrium with r^2>0.95.

References

1 Semel E, Wiig EH, Secord WA: Clinical evaluation of language fundamentals, fourth edition (celf-4). Toronto, The Psychological Corporation/A Harcourt Assessment Company, 2003.
2 Wiig EH, Secord WA, Semel E: Clinical evaluation of language fundamentals—preschool, second edition (celf preschool-2). Toronto, The Psychological Corporation/A Harcourt Assessment Company, 2004.
3 Wechsler D: Wechsler abrreviated scale of intelligence. San Antonio, TX, The Psychological Corporation, 1999,
4 Carrow-Woolfolk E: Comprehensive assessment of spoken language. Circle Pines, MN, AGS, 1999.
5 Wagner RK, Torgesen JK, Rashotte CA: Comprehensive test of phonological processing. Austin, TX, PRO-ED, 1999.
6 Wiederholt JL, Bryant BR: Gray oral reading test-iv. Austin, TX, Pro-Ed, 2001.
7 Woodcock RN: Woodcock reading mastery tests-revised examiners manual. Circle Pines, MN, AGS, 1987.
8 Wilkinson GS: Wide range achievement test-revised 3. Wilmington, DE, Jastak Associates;, 1984.
9 Simmons TR, Flax JF, Azaro MA, Hayter JE, Justice LM, Petrill SA, Bassett AS, Tallal P, Brzustowicz LM, Bartlett CW: Increasing genotype-phenotype model determinism: Application to bivariate reading/language traits and epistatic interactions in language-impaired families. Hum Hered 2010;70:232-244.
10 Abecasis GR, Cherny SS, Cookson WO, Cardon LR: Merlin--rapid analysis of dense genetic maps using sparse gene flow trees. Nat Genet 2002;30:97-101.
11 Purcell S, Neale B, Todd-Brown K, Thomas L, Ferreira MA, Bender D, Maller J, Sklar P, de Bakker PI, Daly MJ, Sham PC: Plink: A tool set for whole-genome association and population-based linkage analyses. Am J Hum Genet 2007;81:559-575.
12 Broman KW, Weber JL: Estimation of pairwise relationships in the presence of genotyping errors. Am J Hum Genet 1998;63:1563-1564.
13 Almasy L, Blangero J: Multipoint quantitative-trait linkage analysis in general pedigrees. Am J Hum Genet 1998;62:1198-1211.
14 Higham N: Computing the nearest correlation matrix - a problem from finance. Journal of Numerical Analysis 2002;22
15 Horn J: A rationale and test for the number of factors in factor analysis. Psychometrika 1965;30
16 Price AL, Patterson NJ, Plenge RM, Weinblatt ME, Shadick NA, Reich D: Principal components analysis corrects for stratification in genome-wide association studies. Nat Genet 2006;38:904-909.
17 Vieland VJ, Huang Y, Seok SC, Burian J, Catalyurek U, O'Connell J, Segre A, Valentine-Cooper W: Kelvin: A software package for rigorous measurement of statistical evidence in human genetics. Hum Hered 2011;72:276-288.
18 Bartlett CW, Vieland VJ: Two novel quantitative trait linkage analysis statistics based on the posterior probability of linkage: Application to the coga families. BMC genetics 2005;6 Suppl 1:S121.
19 Hou L, Wang K, Bartlett CW: Evaluation of a bayesian model-integration-based method for censored data Hum Hered in press

20 International HapMap Consortium: The international hapmap project. Nature 2003;426:789-796.
21 Barrett JC, Fry B, Maller J, Daly MJ: Haploview: Analysis and visualization of ld and haplotype maps. Bioinformatics 2005;21:263-265.