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Association of *MET* with Social and Communication Phenotypes in Individuals with Autism Spectrum Disorder

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Abstract

Autism is a complex neurodevelopmental disorder diagnosed by impairments in social interaction, communication, and behavioral flexibility. Autism is highly heritable, but it is not known whether a genetic risk factor contributes to all three core domains of the disorder or autism results from the confluence of multiple genetic risk factors for each domain. We and others reported previously association of variants in the gene encoding the MET receptor tyrosine kinase in five independent samples. We further described enriched association of the *MET* promoter variant rs1858830 C allele in families with co-occurring autism and gastrointestinal conditions. To test the contribution of this functional *MET* promoter variant to the domains of autism, we analyzed its association with quantitative scores derived from three instruments used to diagnose and describe autism phenotypes: the Autism Diagnostic Interview-Revised (ADI-R), the Autism Diagnostic Observation Schedule (ADOS), and both the parent and the teacher report forms of the Social Responsiveness Scale (SRS). In 748 individuals from 367 families, the transmission of the *MET* C allele from parent-to-child was consistently associated with both social and communication phenotypes of autism. Stratification by gastrointestinal conditions revealed a similar pattern of association with both social and communication phenotypes in 242 individuals with autism from 118 families with co-occurring gastrointestinal conditions, but a lack of association with any domain in 181 individuals from 96 families with ASD and no co-occurring gastrointestinal condition. These data indicate that the *MET* C allele influences at least two of the three domains of the autism triad.

Keywords

HGF; hepatocyte growth factor; gastrointestinal; SRS

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INTRODUCTION

Autism Spectrum Disorder (ASD) is a complex and heterogeneous neurodevelopmental disorder diagnosed by phenotypes in three core symptom domains: (1) verbal and non-verbal communication; (2) social interactions; and (3) behavioral inflexibility (American Psychiatric Association, 2000). Twin studies indicate that the risk for ASD is highly heritable (Bailey et al., 1995; Steffenburg et al., 1989), but genetic linkage studies have failed to identify a single chromosomal region of strong effect (Abrahams and Geschwind, 2008; O'Roak and State, 2008). One possible interpretation of these data is that genetic risk factors may contribute combinatorially to each of the three phenotypic domains of ASD. This line of reasoning suggests that there are genetic variants that influence independently social behavior, language development, and behavioral flexibility, and that ASD results from a confluence of multiple genetic risk factors for each of the three domains. An alternative hypothesis is that the core phenotypes that characterize ASD are mediated through a common biological mechanism, and thus the three domains are not separable. In the latter hypothesis, the observed genetic complexity is due to vulnerability imparted by multiple genes encoding proteins within a common biological pathway.

Principal components analyses (PCA) of the elements that comprise the instruments used to diagnose and describe autism phenotypes have supported both multiple-domain and single-factor hypotheses of ASD. These standardized tests include the Autism Diagnostic Interview-Revised (ADI-R) (Rutter et al., 2003), the Autism Diagnostic Observation Schedule (ADOS) (Lord et al., 1999), and the Social Responsiveness Scale (SRS) (Constantino, 2002). The ADOS and ADI-R are common, standardized instruments that use cut-off scores for clinical diagnosis of autism. The ADOS is a semi-structured, standardized assessment of communication, social interaction, and play or imaginative use of materials. The instrument is administered by a trained clinician to individuals at risk for ASD. There are four modules that are based on the individual's level of expressive language skills. The ADI-R, also used for diagnosis, is an extended interview designed to elicit the parent's/caregiver's history of the child's communication, reciprocal social interactions, and restricted, repetitive, and stereotyped behaviors and interests. The SRS is a continuous rating scale completed by parents or teachers, and is used to assess the severity of social impairments in any population. PCA of the ADI-R and the SRS in a sample of 226 individuals with ASD resulted in a single factor explaining the majority of the variance, supporting the single-factor hypothesis (Constantino et al., 2004). In contrast, PCA of the ADI-R in four independent samples, each including 200–400 individuals with ASD, identified 3–6 factors, supporting the multiple-domain hypothesis (Boomsma et al., 2008; Lecavalier et al., 2006; Tadevosyan-Leyfer et al., 2003; van Lang et al., 2006). In a recent report of a much larger 1,170-individual sample, PCA of the ADI-R resulted in support for a two-factor model, suggesting an intermediate complexity to ASD etiology (Frazier et al., 2008).

One way to distinguish among the multiple-domain and single-factor hypotheses of ASD susceptibility is to test association of ASD-associated genetic variants with phenotype scores on the standardized evaluations of autism traits, the ADI-R and ADOS, and with quantitative traits measured on the SRS (Duvall et al., 2007). If the genetic variant is associated with

some domain scores but not others, then the multiple-domain hypothesis would be supported. In contrast, if the genetic variant was uniformly associated with all domain scores, then the single-factor hypothesis would be supported. This approach has been described for a number of ASD candidate gene studies (Alarcon et al., 2008; Kim et al., 2008a; Kim et al., 2008b; Lerer et al., 2008; Mulder et al., 2005; Sutcliffe et al., 2005; Yrigollen et al., 2008) with mixed results. For example, the serotonin transporter gene (*SLC6A4*) is associated with the compulsive behaviors of individuals with ASD (Mulder et al., 2005; Sutcliffe et al., 2005), a result that is supported by other reports of *SLC6A4* association with obsessive-compulsive disorder (Hu et al., 2006; Wendland et al., 2008). Genetic variants of the *CNTNAP2* gene are associated with delayed language development in individuals with ASD (Alarcon et al., 2008) and with a quantitative language phenotype in individuals with specific language impairment (Vernes et al., 2008). In contrast, analysis of the oxytocin receptor gene (*OXTR*) with ASD phenotypes revealed association with the behavioral inflexibility domain but not the social domain of autism (Lerer et al., 2008; Yrigollen et al., 2008), a finding that is counterintuitive to the known contribution of the oxytocin receptor to the regulation of social behavior (Hammock and Levitt, 2006; Kosfeld et al., 2005).

The relationship between genetic risk and phenotypes characteristic of ASD lies in the patterns of gene expression and the function of the encoded proteins in specific circuits during development. The receptor tyrosine kinase Met is expressed during mouse forebrain development in circuits that are involved in social behavior and emotional regulation (Judson et al., 2009). Based on recent biological studies, we have suggested that MET receptor activation influences the development and maturation of these circuits (Levitt and Campbell, 2009). This neurodevelopmental role is consistent with recent genetic findings of significant association of the *MET* promoter variant rs1858830 C allele with ASD risk in 3 independent cohorts (Campbell et al., 2008; Campbell et al., 2006). An additional report using two cohorts identified significant association of another marker in the *MET* gene with ASD risk (Sousa et al., 2008). Further, *MET* transcript and MET protein are significantly decreased in postmortem brains of individuals with ASD (Campbell et al., 2007). In a recent follow-up study, we described that association of the *MET* rs1858830 C allele was enriched in a subset of individuals with ASD with co-occurring gastrointestinal (GI) conditions (Campbell et al., 2009), which is consistent with disruption of the biological functions of the MET receptor tyrosine kinase in both brain development and GI repair. Here, we tested association of the *MET* rs1858830 C allele with ASD phenotypes measured on the SRS, ADI-R, and ADOS.

METHODS AND MATERIALS

Subjects.

All subjects were collected by the Autism Genetic Resource Exchange (AGRE) Consortium. On February 1, 2008 two data sets, a pedigree file and a medical history file, were downloaded from the AGRE website (www.agre.org). The pedigree file contained genotype information as well as ASD status. Approximately 92% of the families had more than one child with ASD (multiplex). The medical history file contained the child's medical history collected through parent report. The two files were combined to identify children with an

ASD diagnosis and the presence or absence of co-occurring GI symptoms as described previously (Campbell et al., 2009). On January 31, 2009, six additional files were downloaded from the AGRE website: the phenotype scores on the SRS, the ADI-R, and each of the four ADOS modules.

Analyses.

Genotypes at the *MET* promoter variant rs1858830 locus were determined as previously described (Campbell et al., 2009; Campbell et al., 2008; Campbell et al., 2006). No additional families were genotyped for this study. The phenotype scores downloaded from the AGRE website were converted directly to phenotype (.phe) files used for FBAT software analysis. Scores reported here are: (a) quantitative summation scores from individual items on the ADI-R and ADOS; (b) binary cut-off scores from the ADI-R and ADOS; (c) factor scores from a previously published PCA of the AGRE sample (Frazier et al., 2008); and (d) T-scores derived from subscales and total scores on the SRS. We performed PCA of the AGRE ADI-R scores and obtained factor structures that were indistinguishable from those previously reported (Frazier et al., 2008); therefore, we also report association analysis of two factor scores on the ADI-R. Frazier et al (2008) report a 2-factor solution with high loadings of seven components on the first factor (SOC1T_CS, SOC2T_CS, SOC3T_CS, SOC4T_CS, COM1T_CS, COM4T_CS, and COM2VTCS) and three components on the second factor (COM3VTCS, BEH1T_CS, and BEH2T_CS). To determine the probability of obtaining by chance the association of the *MET* promoter variant with co-occurring ASD and GI conditions but not with ASD without GI conditions, we performed 1000 permutations of the 214-family data set into randomly-assigned 118-family and 96-family strata. We report the rank of the observed *P* value for association of the MET rs1858830 C allele with ASD diagnosis among the 1000 *P* values obtained by permutation.

All association analyses were performed with the family-based association test (FBAT) (version 1.7.2; www.biostat.harvard.edu/~fbat) using the empirical variance ("-e") option to account for linkage in the region and to use the observed variance rather than an estimated variance (Horvath et al., 2001). All analyses were performed with both the FBAT additive and dominant models. The additive model represents transmissions from heterozygous parents to offspring, requires 2 degrees of freedom (df), and cannot determine inheritance when two heterozygous parents produce a heterozygous offspring. The dominant model requires 1 df and can determine inheritance of heterozygous offspring (Horvath et al., 2001; Laird and Lange, 2008). The dominant model is appropriate when individuals with one or two risk alleles have the same relative risk (Schaid,1996), as was previously reported for the *MET* rs1858830 C allele in case-control analysis (Campbell et al., 2008; Campbell et al., 2006). For risk alleles, it is expected that the dominant model provides greater power to detect significant differences.

RESULTS

Association of *MET* promoter variant rs1858830 C allele with ASD diagnosis.

We previously reported association of the *MET* promoter variant rs1858830 in three independent cohorts (Campbell et al., 2008; Campbell et al., 2006) consisting of a total of

848 families. In addition to AGRE families, these cohorts also included samples from Italy, Iowa, Stanford, Tufts, and Vanderbilt. In the 367 AGRE families genotyped, FBAT analysis indicated significant association of the *MET* rs1858830 C allele with ASD diagnosis using both the additive model ($P = 0.037$) and the dominant model ($P = 0.0008$) (Table 1). For comparison, the *MET* rs1858830 C allele was similarly associated with ASD diagnosis in 481 non-AGRE families using both the FBAT additive model ($P = 0.033$) and the dominant model ($P = 0.0005$) (Table 1), suggesting that the AGRE sample is representative of the larger 848-family sample. In the combined 848-family sample, the *MET* rs1858830 C allele is strongly associated with ASD diagnosis (additive model $P = 0.003$; dominant model $P = 1 \times 10^{-6}$) (Table 1). In the 214-family sample for which GI report was available, the *MET* rs1858830 C allele was associated with ASD diagnosis in the 118 families with co-occurring ASD and GI conditions (additive model $P = 0.009$; dominant model $P = 0.004$) but was not associated with ASD diagnosis in the 96 families without co-occurring ASD and GI conditions (additive model $P = 0.373$; dominant model $P = 0.205$) (Campbell et al., 2009). Permutation analysis ranked the observed association of the *MET* rs1858830 C allele with co-occurring ASD and GI conditions high among the P values obtained by random stratification (additive model: 103 of 1000; dominant model: 71 of 1000). Given the overall association of the *MET* variant with ASD, we wished to determine whether the *MET* variant was associated with specific phenotypes of the three core domains of ASD measured by the assessment instruments.

Sample and availability of phenotype data.

The sample of 1,699 individuals included 748 diagnosed with ASD, from 367 AGRE families with available phenotype data. Table 2 lists the number of individuals with ASD who were phenotyped using four instruments for measurement of autism traits: (1) the SRS Teacher Report; (2) the SRS Parent Report; (3) the ADI-R; and (4) the ADOS. Table 2 also lists the number of individuals phenotyped with these measures in two subgroups of the total 367 AGRE families: 118 families in which at least one individual with ASD has a co-occurring GI condition and 96 families in which no individual with ASD has a co-occurring GI condition. Phenotype data is available for 153 families for which no indication of GI status is available.

Association of *MET* rs1858830 C allele with SRS Teacher Report phenotype scores.

In the entire 367-family sample, the *MET* rs1858830 C allele was significantly associated with SRS total score by both FBAT additive model ($P = 0.033$) and dominant model ($P = 0.001$) (Table 3). The *MET* C allele was also significantly associated, independent of FBAT model, with each of the five subscales within the SRS for the entire sample (Table 3). In the 118 families with co-occurring ASD and GI conditions, the *MET* C allele was significantly associated with SRS total score by the dominant model ($P = 0.007$) and showed a trend toward association with the additive model ($P = 0.063$) (Table 3). For each of the SRS subscale T-scores in the families with co-occurring ASD and GI conditions, similar results were observed: significant associations with the dominant model and trends toward association with the additive model (Table 3). In the 96 families without co-occurring GI conditions, no significant association was observed for any SRS score (Table 3).

Association of *MET* rs1858830 C allele with SRS Parent Report phenotype scores.

In the entire 367-family sample, the *MET* rs1858830 C allele was significantly associated SRS total score ($P = 0.003$) and each of the SRS subscale scores, but only using the FBAT dominant model (Table 4). In the 118-families with co-occurring ASD and GI conditions, a trend toward association of the *MET* C allele was observed for SRS total score using the FBAT dominant model ($P = 0.067$) and each of the SRS subscale scores (Table 4). However, there was no evidence of association either when using the FBAT additive model or when analyzing families with no co-occurring ASD and GI condition (Table 4).

Association of *MET* rs1858830 C allele with ADI-R phenotype scores.

A. Analysis with FBAT dominant model.—Application of the FBAT dominant model to the entire 367-family AGRE sample revealed association of the *MET* promoter variant rs1858830 C allele with 22 of the 24 ADI-R phenotype scores; both exceptions concerned non-verbal communication (Table 5). The *MET* C allele was associated in the entire sample with quantitative total scores for social ($P = 0.008$), verbal communication ($P = 0.009$), behavior ($P = 0.013$), and abnormal development ($P = 0.003$) phenotypes (Table 5). The *MET* C allele also was associated with binary cut-off scores for autism diagnosis, with each of the phenotype domain cut-off scores except non-verbal communication, and with multiple subscores within each domain (Table 5). In the entire sample, the *MET* C allele also was associated with both of the ADI-R factors defined by PCA of the AGRE family data set (Frazier et al., 2008) (Table 5). A similar pattern of association was observed in the 118 families with co-occurring ASD and GI conditions using the FBAT dominant model (Table 5). The *MET* C allele was associated with quantitative scores for social ($P = 0.020$), verbal communication ($P = 0.011$), and abnormal development ($P = 0.016$), but not with non-verbal communication or behavior in this subset (Table 5). In the subset with co-occurring GI conditions, the *MET* C allele was associated with autism diagnosis cut-off ($P = 0.025$) and with each of the domain cut-off scores except non-verbal communication (Table 5). In the families with co-occurring ASD and GI conditions, the *MET* C allele was associated with Factor 1 of the PCA, which concerns social and communication phenotypes, but not with Factor 2, which includes behavioral inflexibility phenotypes (Frazier et al., 2008) (Table 5). In the 96 families without co-occurring GI conditions, there was no evidence of association of the *MET* C allele with any ADI-R phenotype score (Table 5).

B. Analysis with FBAT additive model.—Association of the *MET* rs1858830 C allele with ADI-R phenotypes was restricted to verbal communication and abnormal development when the FBAT additive model was applied. The *MET* C allele was not associated with the autism cut-off score in any sample using the FBAT additive model (Table 5). In the entire 367-family sample, the *MET* C allele was associated with 5 of 24 phenotypes examined, including quantitative scores for verbal communication ($P = 0.050$), cut-off scores for verbal communication ($P = 0.027$) and abnormal development ($P = 0.040$), and two communication subscales (Table 5). In the subset of 118 families with co-occurring GI conditions, the *MET* C allele was associated with quantitative scores for verbal communication ($P = 0.045$) and abnormal development ($P = 0.047$), and with cut-off scores for verbal communication ($P = 0.025$), communication ($P = 0.037$), and abnormal development ($P = 0.019$) (Table 5). In the

96 families with no co-occurring GI conditions, the *MET* C allele was not associated with any ADI-R phenotype score (Table 5).

Association of *MET* rs1858830 C allele with ADOS cut-off scores.

A total of 528 individuals with ASD were administered the ADOS in the entire 367-family sample. Four distinct modules of the ADOS are administered, depending upon age and verbal abilities of the subject, and it is not appropriate to collapse quantitative phenotype scores across the four ADOS modules. Further, there was not sufficient power in this sample to compare across ADOS modules as the number of subjects in each group ranged from 8 to 214 (mean ± standard deviation = 76 ± 63; data not shown). Therefore, we did not analyze quantitative traits on each of the ADOS modules. However, each ADOS module contains a module-specific algorithm for determining whether an individual meets criteria for autism or autism spectrum disorder, and reports a binary cut-off (yes or no) for meeting the criteria for diagnosis. Analysis of these cut-off scores is presented in Table 6. Using the FBAT dominant model, association of the *MET* rs1858830 C allele is significant for the autism cut-off in the entire sample for communication ($P = 0.008$) and social (P=0.002) phenotypes, and for the social and communication domains combined ($P = 0.005$) (Table 6). In the sample of 118 families with co-occurring GI conditions, the same pattern is observed with the FBAT dominant model: the *MET* C allele is associated with the autism cut-off for communication ($P = 0.029$) and social phenotypes ($P = 0.011$), and for the social and communication domains combined ($P = 0.018$) (Table 6). Application of the FBAT additive model revealed that association of the *MET* C allele is restricted to the autism spectrum cut-off. In the entire sample, the *MET* C allele was only associated with the communication autism spectrum cut-off ($P = 0.048$), but not with the social cut-off or any autism cut-off (Table 6). In the subset of 118 families with co-occurring ASD and GI conditions, the MET C allele was associated with the autism spectrum cut-off for communication ($P = 0.022$) and social ($P = 0.024$) phenotypes, and with the social and communication domains combined ($P = 0.026$), but was not associated with any autism cut-off score. In the sample of 96 families without co-occurring GI conditions, there was no evidence of association of the *MET* C allele with ADOS cut-off scores.

DISCUSSION

The present analysis of the relationship between genetic risk and functional phenotypes, based on scores from diagnostic and analytical instruments, reveals that the *MET* C allele influences at least two of the three domains typically used to characterize ASD. There was consistent association, irrespective of the analytical model used, with social and communication phenotypes. Moreover, the *MET* C allele showed a statistically significant association with ADI-R Factor 1 based on PCA (Frazier et al., 2008), again indicating a broadly applicable increase in risk of multiple phenotypes spanning the social and communication domains. The inclusion of repetitive behaviors occurred using only the dominant model, supporting our hypothesis that the *MET* C allele influences the development of multiple brain circuits that underlie the constellation of complex behaviors. This makes sense biologically given the patterns of MET expression in the developing brain. Although not yet known in the developing human brain, our recent analysis in the mouse

demonstrate that in the forebrain, there is an enrichment of Met expression in projection neurons of the cerebral cortex, hippocampus and amygdala particularly during the critical period of synapse formation and pruning (Judson et al., 2009). These circuits provide top-down control to the striatum, thalamus and subcortical limbic structures, and integration of information across cortical areas. Each of these circuits is involved in core social, communication and behavioral flexibility domains that are disrupted in ASD. Thus, given these expression patterns, risk occurring through the *MET* variant that disrupts gene transcription would be more likely to influence multiple phenotypes rather than a single feature. In fact, our analysis of subdomains of the SRS, ADOS and ADI-R all revealed a consistent influence of the *MET* variant on social and communication features of ASD.

Pleiotropic Influences of Genetic Risk on Multiple Phenotypes in ASD

FBAT additive model analysis of both ADI-R and ADOS suggest that the *MET* C allele is associated with the broader autism spectrum. The *MET* C allele was not associated with either surpassing the narrow autism cut-off score in the ADI-R (Table 5) or surpassing the narrow autism communication and social cut-off score in the ADOS (Table 6) when using the FBAT additive model. Moreover, when a subgroup of the entire sample was analyzed based on presence or absence of GI condition, association of the *MET* C allele with several phenotypic components of the instruments was observed only in the families in which there was co-occurring ASD and GI conditions in at least one child. Although it is possible that the negative findings in the ASD-non-GI families may be hampered in part by insufficient power when stratification was performed, the data from the positive findings are consistent with our hypothesis that a single, key biological pathway may influence multiple neurodevelopmental and medical outcomes. This provides a biological framework upon which principles of brain and peripheral organ development and plasticity related to ASD may be investigated, without the need to implicate a single anatomically-localized dysfunction as the cause for all other phenotypes that characterize individuals with ASD. The data also are consistent with the idea that there will be a combination of genes that provide risk for ASD. We suggest that disorder heterogeneity is reflected in unique combinations of mechanisms that include heritable risk variants, such as the *MET* C allele, *de novo* mutations and copy number variations, and genetic and environmental modifiers of core phenotypes and associated medical conditions. The MET biological pathway, which includes the receptor, its only known ligand hepatocyte growth factor, and the proteins that mediate receptor signaling can be influenced by all of these mechanisms (Campbell et al., 2008; Levitt and Campbell, 2009).

Stratification Analyses Reveal Broad Domains of *MET* Influence

The ADI-R and the ADOS are instruments designed to diagnose the clinical disorder, and thus provide a set of cut-off scores that are not sensitive for revealing quantitative differences between individuals within any domain. The statistical association of the transmission of the *MET* C allele to those individuals above the cut-off for ASD diagnosis in all domains using the dominant model, and two of three using the additive model, indicate that this allele is related to the disorder per se rather than to any single phenotypic feature. The dominant model also provided support for association with the narrow definition of autism, with the additive model revealing association only with the broader spectrum. Support for a disorder-

related global association also is derived from consistent findings across the two diagnostic instruments. Further broad influences of the *MET* C allele are revealed by our finding that the variant is associated with SRS scores, which are designed to describe quantitative features of social and communication phenotypes in the broader population. In order to determine unequivocally that the *MET* allelic variant is related to quantitative features of social and communication functions, rather than the impairments specific to the clinical population with autism, comparison of inheritance patterns in the general population will be necessary.

There were differences in association of the *MET* C allele when the SRS scores were stratified by parent and teacher report. Both showed significant association using the dominant model, but only teacher report scores were significant when using the additive model. There are reported increases in the expression of broader autism phenotypes in multiplex compared to simplex families (Losh et al., 2008; Virkud et al., 2008). Given that our sample was predominantly multiplex families, the differences in associations with the social and communication modules based on parent and teacher scores may reflect differences in the interpretation of questions and the reporting of features using SRS. With respect to differences between the teacher-report and parent-report SRS scores, our results are similar to those observed for linkage signals in the AGRE cohort (Duvall et al., 2007). The linkage signals were stronger across several loci for the teacher-report SRS scores compared to the parent-report SRS scores, prompting Duvall et al (2007) to suggest the possibility of modest rater contrast effects on the part of parents, but not teachers. We also noted that in the subgroup of families with information regarding GI condition, the statistically significant relationship of subscores on the diagnostic instruments and the SRS were found only in those families in which at least one child had co-occurring ASD and a parent-reported GI condition. We reported previously genetic findings of *MET*, *SERPINE1* and *PLAUR* variant association in multiplex, but not simplex families (Campbell et al., 2008; Campbell et al., 2006), and in subgroups only with co-occurring GI conditions related to *MET* (Campbell et al., 2009). We suggest that beyond the analysis of subcomponents of the phenotypes that define the clinical condition, inclusion of unique aspects of family histories may be helpful in generating more precise definitions of ASD etiology that take into account heterogeneity in disorder expression and causes.

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Table 1.

Association of *MET* rs1858830 C allele with ASD diagnosis.

Sample	Pedigrees	FBAT Model	Inf Fams	TOBS	TEXP	Z	P
AGRE Consortium	367						
		Additive	158	377	349	2.081	.037
		Dominant	122	199	174	3.340	.0008
Non-AGRE	481						
(Italy, Iowa, Stanford,		Additive	204	364	339	2.138	.033
Tufts, Vanderbilt)		Dominant	166	186	161	3.449	.0005
Combined	848						
		Additive	362	741	688	2.975	.003
		Dominant	288	385	335	4.830	.000001

Table 2.

Description of phenotypes available for the AGRE sample.

	All Families Genotyped	Families with Co-Occurring GI	Families without Co-Occurring GI
Number of Families	367	118	96
Number of Individuals Phenotyped			
Social Responsiveness Scale (SRS) Teacher Report	294	108	71
Social Responsiveness Scale (SRS) Parent Report	363	123	101
Autism Diagnostic Interview- Revised (ADI-R)	742	242	181
Autism Diagnostic Observation Schedule (ADOS)	528	220	160

Table 3.

Association of *MET* rs1858830 C allele with SRS Teacher Report phenotypes (P values)

	All Families Genotyped		Families with Co- Occurring GI		Families without Co- Occurring GI	
FBAT Model	**Additive**	**Dominant**	**Additive**	**Dominant**	**Additive**	**Dominant**
Trait						
SRS Total	**0.033**	**0.001**	0.063	**0.007**	0.579	0.651
Social Awareness	**0.036**	**0.001**	**0.050**	**0.005**	0.551	0.612
Social Cognition	**0.032**	**0.002**	0.054	**0.008**	0.549	0.660
Social Communication	**0.034**	**0.001**	0.061	**0.007**	0.636	0.693
Social Motivation	**0.029**	**0.001**	0.096	**0.007**	0.606	0.670
Autistic Mannerisms	**0.030**	**0.001**	0.056	**0.007**	0.562	0.613

Table 4.

Association of *MET* rs1858830 C allele with SRS Parent Report phenotypes (P values).

	All Families Genotyped		Families with Co- Occurring GI		Families without Co- Occurring GI	
FBAT Model	**Additive**	**Dominant**	**Additive**	**Dominant**	**Additive**	**Dominant**
Trait						
SRS Total	0.203	**0.003**	0.341	0.067	0.443	0.134
Social Awareness	0.127	**0.002**	0.269	0.080	0.413	0.115
Social Cognition	0.144	**0.001**	0.218	**0.040**	0.408	0.131
Social Communication	0.250	**0.003**	0.392	0.075	0.482	0.130
Social Motivation	0.237	**0.005**	0.434	0.058	0.568	0.272
Autistic Mannerisms	0.174	**0.002**	0.314	0.058	0.382	0.098

Table 5.

Association of the *MET* rs1858830 C allele with phenotypes on the ADI-R (P values)

		All Genotyped Families		Families with Co-Occurring GI		Families without Co-Occurring GI	
	FBAT Model	**Additive**	**Dominant**	**Additive**	**Dominant**	**Additive**	**Dominant**
Variable	**Trait Description**						
SOCT_CS	Social Total	0.231	**0.008**	0.174	**0.020**	0.784	0.679
COMVT_CS	Communication Verbal Total	**0.050**	**0.009**	**0.045**	**0.011**	0.602	0.891
COMNVTCS	Communication Non-Verbal Total	0.891	0.105	0.810	0.421	0.357	0.130
BEHT_CS	Behavior Total	0.323	**0.013**	0.215	0.053	0.565	0.517
DEVT_CS	Abnormality of Development evident at or before 36 months - Total	0.146	**0.003**	**0.047**	**0.016**	0.598	0.331
AUTISMCS	Met ADI Autism cutoff?	0.281	**0.007**	0.231	**0.025**	0.633	0.460
SOCCUTCS	Did child's Social score surpass Autism cutoff?	0.073	**0.001**	0.050	**0.005**	0.549	0.352
CMVCTCS	Did child's Verbal Communication score surpass Autism cutoff?	**0.027**	**0.005**	**0.025**	**0.014**	0.810	0.930
CMNVCTCS	Did child's Non- Verbal Communication score surpass Autism cutoff?	0.963	0.091	0.819	0.437	0.414	0.112
COMCUTCS	Did child's Communication score surpass Autism cutoff?	0.059	**0.001**	**0.037**	**0.013**	0.533	0.327
BEHCUTCS	Did child's Behavior score surpass Autism cutoff?	0.168	**0.002**	0.080	**0.008**	0.385	0.223
DEVCUTCS	Did child's Abnormality of Development score surpass Autism cutoff?	**0.040**	**0.001**	**0.019**	**0.008**	0.412	0.250
SOC1T_CS	Total (Failure to use nonverbal behaviors to regulate social interaction)	0.538	**0.041**	0.497	0.077	0.865	1.000
SOC2T_CS	Total (Failure to develop peer relationships)	0.204	**0.004**	0.166	**0.012**	0.674	0.561
SOC3T_CS	Total (Lack of shared enjoyment)	0.103	**0.002**	0.078	**0.006**	0.625	0.558
SOC4T_CS	Total (Lack of socioemotional reciprocity)	0.270	**0.021**	0.177	**0.043**	0.817	0.708
COM1T_CS	Total for lack of, or delay in, spoken language & failure to compensate through gesture	0.151	**0.006**	0.132	**0.017**	0.201	0.250
COM4T_CS	Total (Lack of varied spontaneous make-believe or social Imitative play)	0.221	**0.005**	0.086	**0.014**	0.891	0.570
COM2VTCS	Total (Relative failure to initiate or sustain conversational interchange)	0.044	**0.025**	0.035	**0.026**	0.687	0.857
COM3VTCS	Total (Stereotyped, repetitive or idiosyncratic speech)	0.023	**0.003**	0.105	**0.043**	0.273	0.507
BEH3T_CS	Stereotyped and repetitive motor mannerisms	0.129	**0.003**	0.285	**0.107**	0.303	0.192

		All Genotyped Families		Families with Co-Occurring GI		Families without Co-Occurring GI	
	FBAT Model	**Additive**	**Dominant**	**Additive**	**Dominant**	**Additive**	**Dominant**
Variable	**Trait Description**						
BEH4T_CS	Preoccupation with part_objects or non-functional elements of materials	0.358	**0.025**	0.050	**0.019**	0.844	0.643
FACTOR1	Factor 1 from Frazier et al, 2008	0.180	**0.006**	0.121	**0.015**	0.662	0.586
FACTOR2	Factor 2 from Frazier et al, 2008	0.082	**0.013**	0.370	**0.191**	0.418	0.598

Table 6.

Association of the *MET* rs1858830 C allele with phenotypes on the ADOS (P values)

		All Genotyped Families		Families with Co-Occurring GI		Families without Co-Occurring GI	
	FBAT Model	**Additive**	**Dominant**	**Additive**	**Dominant**	**Additive**	**Dominant**
Variable	**Trait Description**						
CSACommunication	Communication Autism Cutoff	0.159	**0.008**	0.098	**0.029**	0.460	0.288
CSAS Communication	Communication Autism Spectrum Cutoff	**0.048**	**0.002**	**0.022**	**0.010**	0.332	0.199
CSA Social	Social Autism Cutoff	0.145	**0.002**	0.073	**0.011**	0.455	0.187
CSASSocial	Social Autism Spectrum Cutoff	0.095	**0.001**	**0.024**	**0.009**	0.598	0.217
CSAComSoc	Commmunication + Social Autism Cutoff	0.096	**0.005**	0.058	**0.018**	0.333	0.216
CSASComSoc	Commmunication + Social Autism Spectrum Cutoff	0.113	**0.002**	**0.026**	**0.010**	0.555	0.273