

Comparative Genomics of Autism, Tourette syndrome and Autoimmune/ Inflammatory Disorders 4-23-03

Kevin G. Becker^{1*}, Boris Freidlin², and Richard M. Simon²

¹DNA Array Unit, National Institute on Aging, National Institutes of Health, Baltimore, MD 21224; ²Biometrics Research Branch, National Cancer Institute, Rockville, MD 20892

*Address correspondence to:

Kevin G. Becker, Ph.D.
TRIAD Technology Center
333 Cassell Drive
National Institute on Aging
National Institutes of Health
Baltimore, MD 21224
TEL: 410-558-8360
FAX: 410-558-8281
Email: beckerk@grc.nia.nih.gov

Key words: Autism, Tourette syndrome, Autoimmune

Abbreviations:

autism	Aut
Tourette Syndrome	TS
autoimmune/ inflammatory disorders	AI
multiple sclerosis	MS
systemic lupus erythematosus	SLE
systemic lupus erythematosus with neuropsychiatric phenotype	SLE-NP
Crohn's disease	CD
Psoriasis	PS
Type I diabetes	IDDM
Ankylosing spondylitis	ANK
Obsessive compulsive disorder	OCD
Attention Deficit Hyperactivity Disorder	ADHD

Abstract

Autism and Tourette syndrome are complex enigmatic disorders. Both are childhood neurological syndromes affecting behavior, social interaction, movement, and language. Like many complex disorders, autism and Tourette syndrome are thought to have complex genetic and environmental etiologies. Similarly, autoimmune and inflammatory (AI) disorders have complex multigenic and environmental origins. Genetic loci for AI disorders have previously been shown to cluster non-randomly in several regions throughout the human genome, suggesting in some cases, shared genetic etiologies. Here, we show that genetic loci for autism and Tourette syndrome tend to cluster with each other and also tend to cluster with previously defined loci for inflammatory disorders. This suggests a genetic relationship between autism and Tourette syndrome, as well as a genetic relationship of both with disorders of immune dysregulation.

Introduction

Autism and Tourette syndrome are found in all populations, cultures, and racial groups [1,2]. Both disorders are considered spectrum disorders with a range of clinical symptoms and intermediate phenotypes including individuals with high function to those with profound disability [3,4]. Although both disorders are distinct clinical entities, there are a number of epidemiological and clinical characteristics that are similar between autism and Tourette syndrome including disease prevalence, sex ratio, overlapping traits or intermediate phenotypes, and co-morbidity. The prevalence in the population is similar, with approximately 2-5 cases per 10,000 for autism [2] and approximately 5 per 10,000 for Tourette syndrome [5]. The age of diagnosis for autism is approximately 2-4 years [2] while the age of onset of tics in Tourette syndrome is 2-11 years, with a mean of age of onset of 5.6 years [5]. In both syndromes there is a skewed sex ratio of approximately 4:1 males versus females [2,5]. It has been suggested that the prevalence for both disorders has been increasing, although in both cases it is unclear whether this is a true increase or a change in diagnostic classification and reporting [3,6].

Both disorders were earlier thought to have neuropsychological origins but now both are considered to have complex genetic, environmental, or developmental etiologies [1,2]. In both disorders there is little evidence of degenerative neuropathology, however in both cases subtle anomalies in brain imaging have been observed [7-10]. Autism is characterized primarily by social withdrawal, language impairment, and repetitive behaviors [2], while Tourette syndrome, often grouped with obsessive compulsive disorder (OCD), is generally characterized by vocal and motor tics, behavior problems, impulse control, and compulsive behaviors [1]. Individuals with one disorder sometimes share characteristics of the other, such as verbal and motor tics

being found in some autistic individuals, as well social withdrawal found in some Tourette syndrome or OCD patients [3,11,12]. Both disorders have overlapping clinical characteristics associated with attention deficit hyperactivity disorder (ADHD) [1,5], and co-localization of loci between autism and ADHD has been noted [13]. Co-morbidity for autism and Tourette syndrome has been shown with a dual diagnosis of autism and Tourette syndrome [14-17] within individual patients. Moreover, in both disorders individual clinical characteristics of the respective disorder has been observed in unaffected parents and family members [1,2]. Immune imbalances have been demonstrated in both autism and Tourette syndrome and suggestions of immune or infectious etiology have been made for both disorders [18-21].

Genetics of Autism and Tourette syndrome

The familial nature of both autism and Tourette syndrome has been well established. Through twin studies and family studies, both disorders have been shown to have a significant genetic component with a high heritability estimate of approximately 90% for autism [2,22] and 89-94% for Tourette syndrome [1]. Linkage studies using whole genome scanning approaches have been used in both autism and Tourette syndrome. In both disorders, multiple loci have been identified on different chromosomes suggesting heterogeneity and non-Mendelian inheritance. In most studies, candidate loci are of suggestive significance, rather than confirmed, and in no case has a specific gene been identified for either disorder. As with many genome scans of complex heterogeneous disorders, there are concerns with regard to replication between different studies within a given disorder.

Autism

At least eleven studies have reported whole genome scans of autism or autistic spectrum disorders using sib pair and family studies [4,23-32]. In these reports, no loci of major effect were found, however, all studies found multiple loci of suggestive significance. These loci include positive genome scan results on chromosome 1p, 1q, 2p, 2q, 3p, 3q, 4p, 5p, 6q, 7q, 13q, 15q, 16p, 19p, 19q, Xq, and, Xq-ter, among others (supp.1, supp. 2). Each study analyzed different patient collections, using somewhat different diagnostic criteria, statistical models, and different polymorphic marker panels. Even so, there was replication and overlap of findings between studies (fig 1a-1d; supp. 1), particularly on chromosomes 2q, 7q, 15q, 16p, and 19q. In no case have genes been identified as the source of the genetic linkage.

Tourette syndrome

Likewise, genome-scanning approaches have been used in Tourette syndrome and related disorders such as OCD, by at least 6 groups [33-39]. Also, as before, in most studies no loci of major effect were found, however multiple loci of suggestive significance were identified, including 1p, 2p, 2q, 4p, 4q, 5p, 6p, 7q, 8p, 8q, 11p, 11q, 12p, 12q, 13q, 14q, 16p, 17p, 19p, 20q, and 21q. (supp.1, supp. 2). Replication between different studies has been shown on chromosomes 1p, 2q, 4p, 11q, and 13q.

Immune imbalance in Autism and Tourette syndrome

Both autism and Tourette syndrome have long been suspected to have an immune or infectious component [20,40-43]. The immune component suggested in both disorders is not of a classical autoimmune pattern, such as found in Type I diabetes or multiple sclerosis, with overt

immune cellular infiltration or immune mediated tissue destruction. It may involve subtle antibody mediated effects on brain structures or dysregulation of immune mediators in the context of developmental/ neuroimmune interactions.

Autism has been shown to be associated with cytokine imbalances [44-46] as well as antibody abnormalities including antibodies directed against brain and neuronal factors [47-49]. Infectious agents [50-51], and food allergies [52] have also been suspected to play a role in disease etiology. Autism has been treated with immunological approaches [53]. Interestingly, the frequency of autoimmune disorders has been shown to be increased in families of autistic patients [54] while language impairment and auditory processing deficits have been shown in infants of families with a history of autoimmune disorders [55].

Recently, an association between autism and bowel inflammation has been suggested [56-57] with MMR vaccination in the context of enterocolitis being suggested to play a part in autistic disorder [56,58,59]. The role, if any, of vaccination has been highly debated [60-62], and evidence for this association has not been confirmed in larger studies [63].

Similarly, Tourette syndrome and related disorders have been associated with immune imbalances [18,44,64-66] suggesting an immune component in the etiology of Tourette syndrome. Antibodies have been found in Tourette syndrome patients to phospholipids [67], putamen [68], and other neural and brain epitopes [69]. An association of Tourette syndrome and OCD with streptococcal infections has been demonstrated [70-71] as well. Increased titers of anti-streptococcal antibodies have been found in Tourette syndrome patients [72-73]. These observations and the similarity of Tourette syndrome to the post streptococcal disorder Sydenham's chorea have suggested an antibody mediated or autoimmune model in the etiology of Tourette

syndrome and OCD [74-77]. Rat models of Tourette syndrome using serum from neuronal antibody positive patients resulted in neuronal dysfunction, unprovoked audible vocalizations and increased stereotypies [78-79]. Interestingly, the B-cell marker D8/17 has been found in autism, Tourette syndrome, and OCD patients [80-82].

Paradoxically, none of the whole genome scans that studied autism or Tourette syndrome have found statistically significant linkage to the major histocompatibility complex (MHC) on human chromosome 6p. Recently, Torres and coworkers have demonstrated linkage of the MHC to autism spectrum disorder [83]. Immunological aspects of both autism and Tourette syndrome have been recently reviewed [18,20,42,43,64,78].

Clustering of Loci in Autoimmune / Inflammatory disorders

Classical autoimmune disorders are common chronic conditions of immune dysregulation affecting approximately 4% of the population. These include over 30 diseases such as Type 1 diabetes (IDDM), multiple sclerosis (MS), rheumatoid arthritis (RA), psoriasis (PS), systemic lupus erythematosus (SLE), among many others. Table 1 lists AI disorders described in this study. These disorders involve perturbations in humoral or cellular immune compartments that often lead to chronic immune mediated destruction of organ or tissue targets. In each case, the entire clinical phenotype is largely determined by the extent and functional consequences of immune attack on the target organ. Interestingly, a higher incidence of other autoimmune disorders, but not necessarily the primary disorder, has been demonstrated in families of patients with

specific autoimmune disorders. Likewise, co-morbidity of multiple inflammatory or autoimmune disorders within individuals has been well documented.

The genetics of human autoimmune and inflammatory disorders have long been studied with traditional methods in the context of the major histocompatibility complex (MHC) on chromosome 6p. Many autoimmune or inflammatory disorders have been linked to the MHC through family or sib-pair studies. Recently, autoimmune and inflammatory disorders have been studied using whole genome scanning approaches to identify non-MHC loci. As with other complex disorders, few single genes of major effect have been found in these conditions. However, in each disease, multiple loci of suggestive significance have been found throughout the genome (supp. 1).

It has been shown that MHC and non-MHC loci for many autoimmune and inflammatory disorders linked through whole-genome scanning approaches tend to cluster or co-localize in a statistically significant manner in both mouse and human [84-86]. For example, in addition to the MHC, co-localized clusters (supp. 1) of loci from different autoimmune/ inflammatory disorders can be found at over 30 position in the human genome including; 1p21-22, 1q24-25, 1q42, 2q22, 2q32-36, 3q21, 4q28, 5p15, 5p11, 5q31-33, 6p12-q11, 6q27, 7p15-21, 7q21-22, 7q31, 8q22, 9p22, 10p12, 11p15, 11p13-14, 12p12-13, 14q31-32, 15q11, 15q26, 16q12-21, 17p13, 17q22, 19p13, 19q13, 20p11, 20q13, 21q22, 22q12-13, and Xp11 (supp.1). Clustering of non-MHC loci, has also been shown in animal models of autoimmune and inflammatory disorders including Type I diabetes, systemic lupus erythematosus [84,87], experimental autoimmune encephalomyelitis, experimental autoimmune orchitis [88,89] and arthritis [90].

Explanations for loci clustering may include simple coincidental localization, statistical aberrations, hotspots of recombination in the genome, or uneven distribution of genes and polymorphism in the genome. However, in some cases,

clusters of autoimmune/ inflammatory loci may have a more biologically and clinically relevant basis. For example, the single gene *CTLA4* on chromosome 2q, has been associated with at least seven different autoimmune disorders including, multiple sclerosis, Type I diabetes, Grave's disease, autoimmune hepatitis, celiac disease, rheumatoid arthritis, and Hashimoto's thyroiditis [91,92]. More importantly, overlapping of autoimmune/ inflammatory loci may correspond to dense clusters of evolutionarily conserved functionally related immune genes, such as the MHC, 6p21; cytokine clusters, 5q31-33; NK receptors, 12p12-13; IAN cluster, 7q36; among others [93,94]. Polymorphisms and susceptibility haplotypes within these clusters may not be disease specific, but may perturb basic immune pathways important in multiple conditions [85,91,95].

Results

Comparison of loci in autoimmune/ inflammatory disorders, Autism, and Tourette syndrome.

Diseases compared in this study are shown in table 1. These include all genome scan studies from autistic disorders, Tourette syndrome and related disorders, and a majority of all autoimmune and inflammatory genome scans. In most cases, independent groups have studied the same or similar disorder. Figures 1a-1d are selected composite maps of genetic loci from all three classes of disorders; autism, Tourette syndrome, and autoimmune disorders identified in whole genome scans (see supp. 1, supp. 2 for complete datasets). Data from other types of genetic studies (i.e. chromosomal deletions) are not included. Data from one genome wide scan for ADHD has also been included but

is not included in statistical comparisons. Linkage results shown here are all peak markers of genome scan studies of autism, Tourette syndrome, and autoimmune/inflammatory disorders with significance values of $Lod > 1.0$ or $p\text{-values} < 0.05$. All data above minimum thresholds from all studies are shown. There has been no selection against individual genome scan studies or individual data points, other than as described above. Each linkage is arbitrarily assigned a 10-centimorgan confidence interval position found on the LDB human composite chromosomal map [96]. Actual confidence intervals are quite often unavailable, due to the practice of reporting only peak markers. Ten centimorgans corresponds to the approximate average resolution of genome scans of this type. The thickness of each line is proportional to the significance value (Lod or $p\text{-value}$). All markers, references, and supporting information for all genome studies can be found in the supplementary figures and tables (supp. 1, supp. 2).

As shown in Fig 1a-1d and in the complete dataset (supp 1& 2) loci for all three classes of disorders are non- randomly distributed throughout the genome. This genome wide comparison includes frequent overlapping or coincidence of linkage for all three classes of disorder (autism, Tourette syndrome, & autoimmune). Overlap between linkages for AI and autism include: 1q44, 2q22, 3q21, 6q27, 6p21-p15, 6q11, 7p21-p15, 7cen-q11, 7q31, 9q34, 10p12, 13q12, 13q22-32, 15q11, 15q15-q21, 15q26, 16p13, 19p13, 19cen-q13, 20p13-p12. Coincidence of loci between autoimmune/ inflammatory and Tourette syndrome occurs at: 4p15, 6p25, 6cen, 6q21, 13q14-21, 21q22. Co-occurrence of loci between all three types of disorders is found at: 1p22-p13, 2p16-p13, 2q32-q37, 4q28, 5p15, 5p11-q11, 7q36, 8p23, 8q21-q22, 11p14-p13, 12p12-13, 17p13. Of particular interest are regions where replication has occurred between independent groups within a

single disease type (i.e. autism or Tourette syndrome). The chromosomal region 11q24-25 (fig 1c) has been replicated in four independent studies of Tourette syndrome and has also been linked to autism, SLE and celiac disease. On chromosome 15, autism has been linked to 15q15-q21 in two independent studies, as well as to asthma, and interestingly to dyslexia (supp.1) in three independent studies.

In addition to simple marker co-localization, a striking number of *identical* polymorphic markers have been linked to both autism and autoimmune/ inflammatory disorders or Tourette and autoimmune/ inflammatory disorders. Table 2 summarizes polymorphic markers that have been linked to both autism and AI disorders or Tourette syndrome and AI disorders with *identical* markers. As shown in Table 2, approximately 804 positive markers (Lod > 1.0, p < 0.05) were reported in all autoimmune/ inflammatory genome scan studies described here, 237 markers were positive in autism, and 88 were reported for Tourette/ OCD. Of the 237 autism markers, 89 (37.6%) were found to be statistically significant in both autoimmune disorders and autism. Of a total of 88 markers reported for Tourette/ OCD, a majority, 53 (60.3%), were reported significant for both autoimmune disorders and Tourette syndrome. All positive polymorphic markers (lod > 1.0, p > 0.05) markers for all diseases can be found in supp 2. Identical markers found across disease types can be found in supp 3. Of particular note, given the suggestions of bowel inflammation in autism, are co-localization of markers including *identical* markers linked to both autism and coeliac disease or autism and Crohn's disease.

Genome wide statistical analysis of Autism, Tourette syndrome and Inflammatory/Autoimmune disorders

A randomization test was used to determine if co-localization of loci for autism and Tourette syndrome, autism and AI disorders, and Tourette syndrome and AI disorders could be shown to be statistically significant on a genome-wide scale. For each of the three disease categories autism, Tourette syndrome, and AI disorders, unavailable disease locus confidence intervals were approximated by merging the disease loci that are within 10 cM into contiguous intervals (CI). Significance of clustering of the diseases was evaluated by a randomization test as follows. For each pair of diseases categories, CIs for the disease with fewer studies were randomly repositioned on a 10cM grid covering the human genome, while the CIs for the disease with larger number of studies were kept fixed. This was done because the large number of studies made it difficult to determine which markers identify independent regions. After each repositioning the number of independent CIs was calculated as the number of randomly repositioned CIs of the first disease that are not within 10cM of a CI from the second (fixed) disease. The p-value was taken to be the proportion of the replications resulting in a number of independent intervals no greater than that in the observed data (based on 10,000 replications). As previously described [85], all data from the short arm of chromosome 6 was excluded from statistical analysis due to the strong effect of the MHC on autoimmune disorders.

As shown in Table 3, co-localization of Tourette syndrome loci with AI loci was found to be statistically significant on a genome-wide scale ($P = 0.01$). Co-localization of

Tourette syndrome and autism loci approached, but did not reach, statistical significance ($P = 0.062$). Co-localization of autism and AI disorders could not be determined to be statistically significant on a genome wide scale. However, we believe that this does not preclude description of overlap of loci between autism and AI disorders on an individual locus-by-locus basis.

Discussion

The genetic basis of autism and Tourette syndrome, as well as the genetic components of autoimmune and inflammatory disorders are unclear. Whole genome scans suggest that these disorders are not due to single genes of large effect, but may be influenced by multiple genes, each of moderate to low effect, influencing disease susceptibility. The increasing availability of genomic scale information has allowed novel comparisons across species and across disease with the goal of gaining insight into genetic organization and its relationship to disease.

Here we show a cross disease comparison of candidate loci from autism, Tourette syndrome and multiple autoimmune/inflammatory disorders and the comparative genomic positions of polymorphism data from each disorder. In numerous cases, loci identified in genome scans cluster or co-localize between autism and Tourette syndrome. Moreover, loci found for both autism and Tourette syndrome co-localize with loci identified in autoimmune disorders at multiple locations in the genome. In particular, this clustering quite often occurs in genomic locations, which have been replicated by independent groups within a given disease (ex. 1p22.1-21.2 [TS, Aut], 2p11.2-11.1[TS], 2q32.1-32.3 [Aut], 5p15.3 [Aut], 7p21.2 [Aut], 11q24.1 [TS], 13q14.13-q21.1 [TS]).

Additionally, within this simple marker co-localization, the observation that linkage of a high percentage of *identical* polymorphic markers between autism and autoimmune disorders or Tourette syndrome and autoimmune disorders provides evidence that simple co-localization may not be coincidental.

Overlapping or co-localization of Autism and Tourette syndrome loci alone is interesting given overlapping clinical characteristics between autism and Tourette syndrome, suggesting a shared genetic basis between these neurodevelopmental disorders of language and movement. Co-localization of Autism and Tourette syndrome loci with loci of disorders of immune dysregulation is consistent with a hypothesis of a relationship between immune phenomena and both autism and Tourette syndrome. While there is little evidence of classical immune mediated tissue destruction in either autism or Tourette syndrome, there is evidence of immune imbalance in both disorders. Also, co-localization of linkage between autism and inflammatory gut disorders such as Crohn's disease and celiac disease, allows a testable genetic approach in teasing out a potential relationship between autism and enterocolitis. Of additional interest is linkage of autism and Tourette syndrome to 4p16.3-16.1. This region was recently genetically linked to SLE patients having neuropsychiatric manifestations that, like Tourette syndrome [67], have been associated with antiphospholipid antibodies [97].

Immune influence on neuronal development is well established. Families of cytokines, chemokines, signal transduction molecules, molecules of cell-cell contact, as well as developmental regulatory molecules originally characterized in the immune system have pleiotropic and overlapping functional effects on the developing immune system and the developing brain. Similarly, considerable overlap has been noted between

hematopoetic and neuronal stem cell lineages. In addition, prenatal or neonatal infection has been shown to have a profound influence on brain development with neonatal infection used as a model for autism [98,99].

Co-localization of genetic loci between these clinically distinct disorders suggests a genetic relatedness between all three types of diseases and suggests an underlying genetic basis for immune pathological mechanisms in the etiology of both autism and Tourette syndrome.

Figures

Fig 1. Selected clusters of linkage data from Autism, Tourette syndrome, and Autoimmune/ Inflammatory disorders.

All polymorphic markers are positioned on a common reference map based on LDB gmaps. Chromosome band and centimorgan position are shown above and below respectively for each chromosomal region. All markers are arbitrarily assigned a 10-centimorgan interval at the peak marker reported. Line weight is proportional to LOD score of p-value.

Acknowledgements

The authors would like to thank Drs. Ann Comi, Harvey Singer, John Hardy, and David Schlessinger for critical reading of the manuscript.

Electronic-Database Information

LDB gmaps http://cedar.genetics.soton.ac.uk/public_html/gmap.html

Supplement 1: Composite Genetic Maps For Autism, Tourette syndrome and Autoimmune/Inflammatory Disorders-MAPS

<http://www.grc.nia.nih.gov/branches/rrb/dna/atmap.htm>

Supplement 2: Comparative Genomics of Autism, Tourette syndrome and Autoimmune-Inflammatory Disorders- DATA

<https://www.quickbase.com/db/8jp3dz49>

(Note to reviewers: This is a temporary database. It will become permanent depending on the final journal requirements-KGB)

Supplement 3: Polymorphic markers found in multiple classes of disorders.

<https://www.quickbase.com/db/8qsiujvy>

(Note to reviewers: This is a temporary database. It will become permanent depending on the final journal requirements-KGB)

References

- [1] Jankovic J Tourette's syndrome. *N Engl J Med* 2001 **345**:1184-1192
- [2] Folstein SE, Rosen-Sheidley B. Genetics of autism: complex aetiology for a heterogeneous disorder. *Nat Rev Genet* 2001 **2**:943-955
- [3] Kurlan R, Como PG, Miller B, Palumbo D, Deeley C, Andresen EM, Eapen S, McDermott MP. The behavioral spectrum of tic disorders: A community-based study. *Neurology* 2002 **59**:414-20
- [4] Auranen M, Vanhala R, Varilo T, Ayers K, Kempas E, Ylisaukko-Oja T, Sinsheimer JS, Peltonen L, Jarvela I I. A Genomewide Screen for Autism-Spectrum Disorders: Evidence for a Major Susceptibility Locus on Chromosome 3q25-27. *Am J Hum Genet* 2002 **71**:777-790
- [5] Robertson MM. Tourette syndrome, associated conditions and the complexities of treatment. *Brain* 2000 **123**:425-462
- [6] Gillberg C, Wing L. Autism: not an extremely rare disorder. *Acta Psychiatr Scand* 1999 **99**:399-406
- [7] Peterson BS, Riddle MA, Cohen DJ, *et al* Reduced basal ganglia volumes in Tourette's syndrome using three-dimensional reconstruction techniques from magnetic resonance images. *Neurology* 1993 **43**:941-949
- [8] Singer HS, Reiss AL, Brown RN, *et al*. Volumetric MRI changes in basal ganglia of children with Tourette's syndrome. *Neurology* 1993 **43**:950-956
- [9] Fredericksen KA, Cutting LE, Kates WR, Mostofsky SH, Singer HS, Cooper KL, Lanham DC, Denckla MB, Kaufmann WE. Disproportionate increases of white matter in right frontal lobe in Tourette syndrome. *Neurology* 2002 **58**:85-89
- [10] Sparks BF, Friedman SD, Shaw DW, Aylward EH, Echelard D, Artru AA, Maravilla KR, Giedd JN, Munson J, Dawson G, Dager SR. Brain structural abnormalities in young children with autism spectrum disorder. *Neurology* 2002 **59**:184-192
- [11] Rapin I. Autism spectrum disorders: relevance to Tourette syndrome. *Adv Neurol* 2001 **85**:89-101

- [12] Bejerot S, Nylander L, Lindstrom E. Autistic traits in obsessive-compulsive disorder. *Nord J Psychiatry* 2001 **55**:169-176
- [13] Smalley SL, Kustanovich V, Minassian SL, Stone JL, Ogdie MN, McGough JJ, McCracken JT, MacPhie IL, Francks C, Fisher SE, Cantor RM, Monaco AP, Nelson SF. (2002) Genetic Linkage of Attention-Deficit/Hyperactivity Disorder on Chromosome 16p13, in a Region Implicated in Autism. *Am J Hum Genet* 2002 **71**:959-963
- [14] Sverd J. Tourette syndrome and autistic disorder: a significant relationship *Am J Med Genet* 1991 **39**:173-179
- [15] Baron-Cohen S, Mortimore C, Moriarty J, Izaguirre J, Robertson M. The prevalence of Gilles de la Tourette's syndrome in children and adolescents with autism. *J Child Psychol Psychiatry* 1999 **40**:213-218
- [16] Baron-Cohen S, Scahill VL, Izaguirre J, Hornsey H, Robertson MM. The prevalence of Gilles de la Tourette syndrome in children and adolescents with autism: a large scale study. *Psychol Med* 1999 **29**:1151-1159
- [17] Zappella M. Early-onset Tourette syndrome with reversible autistic behaviour: a dysmaturational disorder. *Eur Child Adolesc Psychiatry* 2002 **11**:18-23
- [18] Hoekstra PJ, Kallenberg CG, Korf J, Minderaa RB. Is Tourette's syndrome an autoimmune disease? *Mol Psychiatry* 2002 **7**:437-445
- [19] Hornig M, Lipkin WI. Infectious and immune factors in the pathogenesis of neurodevelopmental disorders: epidemiology, hypotheses, and animal models. *Ment Retard Dev Disabil Res Rev* 2001 **7**:200-210
- [20] van Gent T, Heijnen CJ, Treffers PD. Autism and the immune system. *J Child Psychol Psychiatry* 1997 **38**:337-349
- [21] Krause I, He XS, Gershwin ME, Shoenfeld Y. Brief report: immune factors in autism: a critical review. *J Autism Dev Disord* 2002 **32**:337-345
- [22] Bailey A, Le Couteur A, Gottesman I, Bolton P, Simonoff E, Yuzda E, Rutter M. Autism as a strongly genetic disorder: evidence from a British twin study. *Psychol Med* 1995 **25**:63-77

- [23] Risch N, Spiker D, Lotspeich L, Nouri N, Hinds D, Hallmayer J, Kalaydjieva L, *et al.* A genomic screen of autism: evidence for a multilocus etiology. *Am J Hum Genet* 1999 **65**:493-507
- [24] International Molecular Genetic Study of Autism Consortium. A full genome screen for autism with evidence for linkage to a region on chromosome 7q. International Molecular Genetic Study of Autism Consortium. *Hum Mol Genet* 1998 **7**:571-578
- [25] Philippe A, Martinez M, Guilloud-Bataille M, Gillberg C, Rastam M, Sponheim E, Coleman M, Zappella M, Aschauer H, Van Maldergem L, Penet C, Feingold J, Brice A, Leboyer M, van Malldergerme Genome-wide scan for autism susceptibility genes. Paris Autism Research International Sibpair Study. *Hum Mol Genet* 1999 **8**:805-812
- [26] Barrett S, Beck JC, Bernier R, Bisson E, Braun TA, Casavant TL, Childress D, *et al.* An autosomal genomic screen for autism. Collaborative linkage study of autism. *Am J Med Genet* 1999 **88**:609-615
- [27] Ashley-Koch A, Wolpert CM, Menold MM, Zaeem L, Basu S, Donnelly SL, Ravan SA, Powell CM, Qumsiyeh MB, Aylsworth AS, Vance JM, Gilbert JR, Wright HH, Abramson RK, DeLong GR, Cuccaro ML, Pericak-Vance MA. Genetic studies of autistic disorder and chromosome 7. *Genomics* 1999 **61**:227-236
- [28] Auranen M, Nieminen T, Majuri S, Vanhala R, Peltonen L, Jarvela I. Analysis of autism susceptibility gene loci on chromosomes 1p, 4p, 6q, 7q, 13q, 15q, 16p, 17q, 19q and 22q in Finnish multiplex families. *Mol Psychiatry* 2000 **5**:320-322
- [29] Cook EH Jr, Courchesne RY, Cox NJ, Lord C, Gonen D, Guter SJ, Lincoln A, Nix K, Haas R, Leventhal BL, Courchesne E. Linkage-disequilibrium mapping of autistic disorder, with 15q11-13 markers. *Am J Hum Genet* 1998 **62**:1077-1083
- [30] Buxbaum JD, Silverman JM, Smith CJ, Kilifarski M, Reichert J, Hollander E, Lawlor BA, Fitzgerald M, Greenberg DA, Davis KL. Evidence for a susceptibility gene for autism on chromosome 2 and for genetic heterogeneity. *Am J Hum Genet* 2001 **68**:1514-1520
- [31] Shao Y, Wolpert CM, Raiford KL, Menold MM, Donnelly SL, Ravan SA, Bass MP, McClain C, von Wendt L, Vance JM, Abramson RH, Wright HH, Ashley-Koch A, Gilbert JR, DeLong RG, Cuccaro ML, Pericak-Vance MA. Genomic screen and follow-up analysis for autistic disorder. *Am J Med Genet* 2002 **114**:99-105
- [32] Liu J, Nyholt DR, Magnussen P, Parano E, Pavone P, Geschwind D, Lord C, Iversen P, Hoh J, Ott J, Gilliam TC The Autism Genetic Resource Exchange

Consortium. A genomewide screen for autism susceptibility loci. *Am J Hum Genet* 2001 **69**:327-340

[33] Barr CL, Wigg KG, Pakstis AJ, Kurlan R, Pauls D, Kidd KK, Tsui LC, Sandor P. Genome scan for linkage to Gilles de la Tourette syndrome. *Am J Med Genet* 1999 **88**:437-445

[34] Simonic I, Gericke GS, Ott J, Weber JL. Identification of genetic markers associated with Gilles de la Tourette syndrome in an Afrikaner population. *Am J Hum Genet* 1998 **63**:839-846

[35] The Tourette Syndrome Association International Consortium for Genetics. A complete genome screen in sib pairs affected by Gilles de la Tourette syndrome. *Am J Hum Genet* 1999 **65**:1428-1436

[36] Merette C, Brassard A, Potvin A, Bouvier H, Rousseau F, Emond C, Bissonnette L, Roy MA, Maziade M, Ott J, Caron C. Significant linkage for Tourette syndrome in a large French Canadian family. *Am J Hum Genet* 2000 **67**:1008-13

[37] Simonic I, Nyholt DR, Gericke GS, Gordon D, Matsumoto N, Ledbetter DH, Ott J, Weber JL (2001) Further evidence for linkage of Gilles de la Tourette syndrome (GTS) susceptibility loci on chromosomes 2p11, 8q22 and 11q23-24 in South African Afrikaners. *Am J Med Genet* 2001 **105**:163-167

[38] Zhang H, Leckman JF, Pauls DL, Tsai CP, Kidd KK, Campos MR; Tourette Syndrome Association International Consortium for Genetics. Genomewide scan of hoarding in sib pairs in which both sibs have Gilles de la Tourette syndrome. *Am J Hum Genet* 2002 **70**:896-904

[39] Hanna GL, Veenstra-VanderWeele J, Cox NJ, Boehnke M, Himle JA, Curtis GC, Leventhal BL, Cook EH Jr. Genome-wide linkage analysis of families with obsessive-compulsive disorder ascertained through pediatric probands. *Am J Med Genet* 2002 **114**:541-552

[40] Warren RP, Margaretten NC, Pace NC, Foster A. Immune abnormalities in patients with autism. *J Autism Dev Disord* 1986 **16**:189-197

[41] Menage P, Thibault G, Martineau J, Herault J, Muh JP, Barthelemy C, Lelord G, Bardos P. An IgE mechanism in autistic hypersensitivity? *Biol Psychiatry* 1992 **31**:210-212

- [42] Trifiletti RR, Packard AM. Immune mechanisms in pediatric neuropsychiatric disorders. Tourette's syndrome, OCD, and PANDAS. *Child Adolesc Psychiatr Clin N Am* 1999 **8**:767-775
- [43] Korvatska E, Van de Water J, Anders TF, Gershwin ME. Genetic and immunologic considerations in autism. *Neurobiol Dis* 2002 **9**:107-125
- [44] Jyonouchi H, Sun S, Le H. Proinflammatory and regulatory cytokine production associated with innate and adaptive immune responses in children with autism spectrum disorders and developmental regression. *J Neuroimmunol* 2001 **120**:170-179
- [45] Croonenberghs J, Bosmans E, Deboutte D, Kenis G, Maes M. Activation of the inflammatory response system in autism. *Neuropsychobiology* 2002 **45**:1-6
- [46] Gupta S, Aggarwal S, Rathanravan B, Lee T. Th1- and Th2-like cytokines in CD4+ and CD8+ T cells in autism. *J Neuroimmunol* 1998 **85**:106-109
- [47] Connolly AM, Chez MG, Pestronk A, Arnold ST, Mehta S, Deuel RK. Serum autoantibodies to brain in Landau-Kleffner variant, autism, and other neurologic disorders. *J Pediatr* 1999 **134**:607-613
- [48] Singh VK, Singh EA, Warren RP. Hyperserotoninemia and serotonin receptor antibodies in children with autism but not mental retardation. *Biol Psychiatry* 1997 **41**:753-755
- [49] Croonenberghs J, Wauters A, Devreese K, Verkerk R, Scharpe S, Bosmans E, Eged B, Deboutte D, Maes M. Increased serum albumin, gamma globulin, immunoglobulin IgG, and IgG2 and IgG4 in autism. *Psychol Med* 2002 **32**:1457-1463
- [50] Singh VK, Lin SX, Yang VC. Serological association of measles virus and human herpesvirus-6 with brain autoantibodies in autism. *Clin Immunol Immunopathol* 1998 **89**:105-108
- [51] Anlar B, Oktem F, Torok T. Human parvovirus B19 antibodies in infantile autism. *J Child Neurol* 1994 **9**:104-105
- [52] Lucarelli S, Frediani T, Zingoni AM, Ferruzzi F, Giardini O, Quintieri F, Barbato M, D'Eufemia P, Cardi E. Food allergy and infantile autism. *Panminerva Med* 1995 **37**:137-141
- [53] Gupta S. Immunological treatments for autism. *J Autism Dev Disord* 2000 **30**:475-479

- [54] Comi AM, Zimmerman AW, Frye VH, Law PA, Peeden JN. Familial clustering of autoimmune disorders and evaluation of medical risk factors in autism. *J Child Neurol* 1999 **14**:388-394
- [55] Benasich AA. Impaired processing of brief, rapidly presented auditory cues in infants with a family history of autoimmune disorder. *Dev Neuropsychol* 2002 **22**:351-372
- [56] Wakefield AJ, Anthony A, Murch SH, Thomson M, Montgomery SM, Davies S, O'Leary JJ, Berelowitz M, Walker-Smith JA. Enterocolitis in children with developmental disorders. *Am J Gastroenterol* 2000 **95**:2285-2295
- [57] Furlano RI, Anthony A, Day R, Brown A, McGarvey L, Thomson MA, Davies SE, Berelowitz M, Forbes A, Wakefield AJ, Walker-Smith JA, Murch SH. Colonic CD8 and gamma delta T-cell infiltration with epithelial damage in children with autism. *J Pediatr* 2001 **138**:366-372
- [58] Wakefield AJ. Enterocolitis, autism and measles virus. *Mol Psychiatry* 2002 **7**:S44-46
- [59] Singh VK, Lin SX, Newell E, Nelson C. Abnormal measles-mumps-rubella antibodies and CNS autoimmunity in children with autism. *J Biomed Sci* 2002 **9**:359-364
- [60] Pavone L, Fiumara A, Bottaro G, Mazzone D, Coleman M. Autism and celiac disease: failure to validate the hypothesis that a link might exist. *Biol Psychiatry* 1997 **42**:72-75
- [61] Walker-Smith J. Autism, bowel inflammation, and measles. *Lancet* 2002 **359**:705-706
- [62] Afzal MA, Minor PD. Vaccines, Crohn's disease and autism. *Mol Psychiatry* 2002 **7**:S49-50
- [63] Taylor B, Lingam R, Simmons A, Stowe J, Miller E, Andrews N. Autism and MMR vaccination in North London; no causal relationship. *Mol Psychiatry* 2002 **7**:S7-8
- [64] Visvanathan K, Trifiletti RR, Altemus M, Zabriskie JB. Autoimmune mechanisms in movement disorders. *Semin Pediatr Neurol* 2000 **7**:103-107
- [65] Marazziti D, Presta S, Pfanner C, Gemignani A, Rossi A, Sbrana S, Rocchi V, Ambrogi F, Cassano GB. Immunological alterations in adult obsessive-compulsive disorder. *Biol Psychiatry* 1999 **46**:810-814

- [66] Kurlan R. Could Tourette syndrome be a neurologic manifestation of rheumatic fever? *Adv Neurol* 2001 **85**:307-10
- [67] Singer HS, Krumholz A, Giuliano J, Kiessling LS. Antiphospholipid antibodies: an epiphenomenon in Tourette syndrome. *Mov Disord* 1997 **12**:738-742
- [68] Singer HS, Giuliano JD, Hansen BH, Hallett JJ, Laurino JP, Benson M, Kiessling LS. Antibodies against human putamen in children with Tourette syndrome. *Neurology* 1998 **50**:1618-1624
- [69] Morshed SA, Parveen S, Leckman JF, Mercadante MT, Bittencourt Kiss MH, Miguel EC, Arman A, Yazgan Y, Fujii T, Paul S, Peterson BS, Zhang H, King RA, Scahill L, Lombroso PJ. Antibodies against neural, nuclear, cytoskeletal, and streptococcal epitopes in children and adults with Tourette's syndrome, Sydenham's chorea, and autoimmune disorders. *Biol Psychiatry* 2001 **50**:566-577
- [70] Kerbeshian J, Burd L, Pettit R. A possible post-streptococcal movement disorder with chorea and tics. *Dev Med Child Neurol* 1990 **32**:642-644
- [71] Allen AJ, Leonard HL, Swedo SE. Case study: a new infection-triggered, autoimmune subtype of pediatric OCD and Tourette's syndrome. *J Am Acad Child Adolesc Psychiatry* 1995 **34**:307-311
- [72] Perlmutter SJ, Garvey MA, Castellanos X, Mittleman BB, Giedd J, Rapoport JL, Swedo SE. A case of pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections. *Am J Psychiatry* 1998 **155**:1592-1598
- [73] Muller N, Riedel M, Straube A, Gunther W, Wilske B. Increased anti-streptococcal antibodies in patients with Tourette's syndrome. *Psychiatry Res* 2000 **94**:43-49
- [74] Kiessling L, Marcotte A, Culpepper L. Antineuronal antibodies in movement disorders. *Pediatrics* 1993 **92**:39-43
- [75] Swedo SE. Sydenham's chorea: a model for childhood autoimmune neuropsychiatric disorders (clinical conference). *JAMA* 1994 **272**:1788-1791
- [76] Swedo SE, Leonard HL, Kiessling L. Speculations on antineuronal antibody-mediated neuropsychiatric disorders of childhood. *Pediatrics* 1994 **93**:323-326
- [77] Murphy TK, Petitto JM, Voeller KK, Goodman WK. Obsessive compulsive disorder: is there an association with childhood streptococcal infections and altered immune function? *Semin Clin Neuropsychiatry* 2001 **6**:266-276

- [78] Hallett J, Kiessling L. Genetics of childhood disorders: XXXV. Autoimmune disorders, part 8: animal models for noninflammatory autoimmune disorders of the brain. *J Am Acad Child Adolesc Psychiatry* 2002 **41**:223-225
- [79] Taylor JR, Morshed SA, Parveen S, Mercadante MT, Scahill L, Peterson BS, King RA, Leckman JF, Lombroso PJ. An animal model of Tourette's syndrome. *Am J Psychiatry* 2002 **159**:657-660
- [80] Swedo SE, Leonard HL, Mittleman BB, Allen AJ, Rapoport JL, Dow SP, Kanter ME, Chapman F, Zabriskie J. Identification of children with pediatric autoimmune neuropsychiatric disorders associated with streptococcal infections by a marker associated with rheumatic fever. *Am J Psychiatry* 1997 **154**:110-112
- [81] Hollander E, DelGiudice-Asch G, Simon L, Schmeidler J, Cartwright C, DeCaria CM, Kwon J, Cunningham-Rundles C, Chapman F, Zabriskie JB. B lymphocyte antigen D8/17 and repetitive behaviors in autism. *Am J Psychiatry* 1999 **156**:317-320
- [82] Murphy TK, Goodman WK, Fudge MW, Williams RC Jr, Ayoub EM, Dalal M, Lewis MH, Zabriskie JB. B lymphocyte antigen D8/17: a peripheral marker for childhood-onset obsessive-compulsive disorder and Tourette's syndrome? *Am J Psychiatry* 1997 **154**:402-407
- [83] Torres AR, Maciulis A, Stubbs EG, Cutler A, Odell D The transmission disequilibrium test suggests that HLA-DR4 and DR13 are linked to autism spectrum disorder. *Hum Immunol* 2002 **63**:311-316
- [84] Vyse TJ, Todd JA. Genetic analysis of autoimmune disease. *Cell* 1996 **85**:311-8
- [85] Becker KG, *et al.* Clustering of non-major histocompatibility complex susceptibility candidate loci in human autoimmune diseases. *Proc Natl Acad Sci U S A* 1998 **95**:9979-9984.
- [86] Wanstrat A, Wakeland E The genetics of complex autoimmune diseases: non-MHC susceptibility genes. *Nat Immunol* 2001 **2**:802-809
- [87] Nguyen C, Limaye N, Wakeland EK. Susceptibility genes in the pathogenesis of murine lupus. *Arthritis Res* 2002 **4**:S255-63
- [88] Teuscher C, Hickey WF, Grafer CM, Tung KS. A common immunoregulatory locus controls susceptibility to actively induced experimental allergic encephalomyelitis and experimental allergic orchitis in BALB/c mice. *J Immunol* 1998 **160**:2751-2756
- [89] Ma RZ, Gao J, Meeker ND, Fillmore PD, Tung KS, Watanabe T, Zachary JF, Offner H, Blankenhorn EP, Teuscher C. Identification of Bphs, an autoimmune disease locus, as histamine receptor H1. *Science* 2002 **297**:620-623

- [90] Griffiths MM, Remmers EF. Genetic analysis of collagen-induced arthritis in rats: a polygenic model for rheumatoid arthritis predicts a common framework of cross-species inflammatory/autoimmune disease loci. *Immunol Rev* 2001 **184**:172-183
- [91] Becker KG. The common genetic hypothesis of autoimmune/inflammatory disease. *Curr Opin Allergy Clin Immunol* 2001 **1**:399-405
- [92] Einarsdottir E *et al.* The CTLA4 region as a general autoimmunity factor: An extended pedigree provides evidence for synergy with the HLA locus in the etiology of type 1 diabetes mellitus, Hashimoto's thyroiditis and Graves' disease. *Eur J Hum Genet* 2003 **11**:81-84
- [93] Flajnik MF and Kasahara M. Comparative genomics of the MHC: glimpses into the evolution of the adaptive immune system. *Immunity* 2001 **15**:351-362
- [94] Trowsdale J. Genetic and functional relationships between MHC and NK receptor genes. *Immunity* 2001 **15**:363-374
- [95] Marrack P, Kappler J, Kotzin BL. Autoimmune disease: why and where it occurs. *Nat Med* 2001 **7**:899-905
- [96] Collins A., Frezal J., Teague J. & Morton NE. A metric map of humans:23,500 loci in 850 bands. *Proc. Natl. Acad. Sci. USA* 1996 **93**:14771-14775.
- [97] Mok CC, Lau CS, Wong RW. Neuropsychiatric manifestations and their clinical associations in southern Chinese patients with systemic lupus erythematosus. *J Rheumatol* 2001 **28**:766-771
- [98] Hornig M, Weissenbock H, Horscroft N, Lipkin WI. An infection-based model of neurodevelopmental damage. *Proc Natl Acad Sci U S A* 1999 **96**:12102-12107
- [99] Patterson PH. Maternal infection: window on neuroimmune interactions in fetal brain development and mental illness. *Curr Opin Neurobiol* 2002 **12**:115-118

Table 1. Diseases Compared

Abbreviations on Figures	Diseases	# of Genome Scans
Aut	Autism	12
Tou	Tourette syndrome	6
OCD	Obsessive compulsive disorder	1
ADHD	Attention deficit hyperactivity disorder	1
DYS	Dyslexia	7
AR	Allergic rhinitis	1
ANK	Ankylosing spondylitis	1
AS	Asthma	8
AD	Atopic dermatitis	1
Celiac	Celiac disease	8
COPD	Chronic obstructive pulmonary disease	1
CD	Crohn's disease	10
GD	Graves disease	4
IBD	Inflammatory bowel disease	1
MS	Multiple sclerosis	8
PS	Psoriasis	6
RA	Rheumatoid arthritis	4
SLE	Systemic Lupus Erythematosus	8
SLE-NP	Systemic Lupus Erythematosus-NP	1
IDDM	Type I diabetes	12

Table 2. Polymorphic markers found positive between autoimmune and autism or autoimmune and Tourette

Disease Group	Total positive markers	Shared with AI
Autoimmune	804	-
Autism	237	89 (37.6%)
Tourette/OCD	88	53 (60.3%)

The complete dataset can be found as supplement # 3
<https://www.quickbase.com/db/8qsiujvy?nt=1>

**Table 3. Statistical association of
all three classes of disease**

Tourette vs autoimmune $P = \mathbf{0.010}$

Tourette vs Autism $P = 0.062$

Autism vs autoimmune $P = 0.311$

P-values are based on 10,000 replications

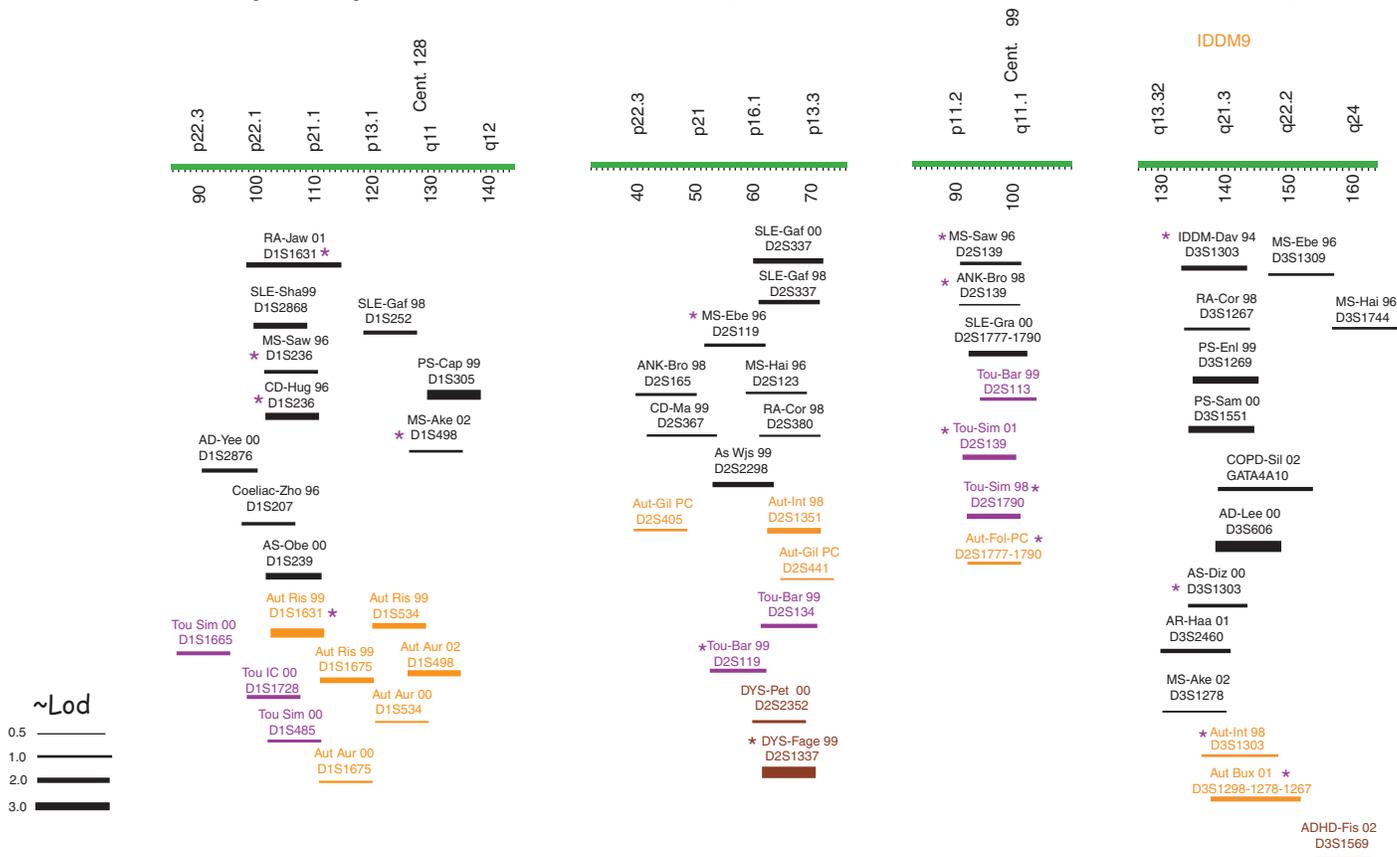
1a

1p22-q12

2p16-p13

2p11-q11

3q21-q22



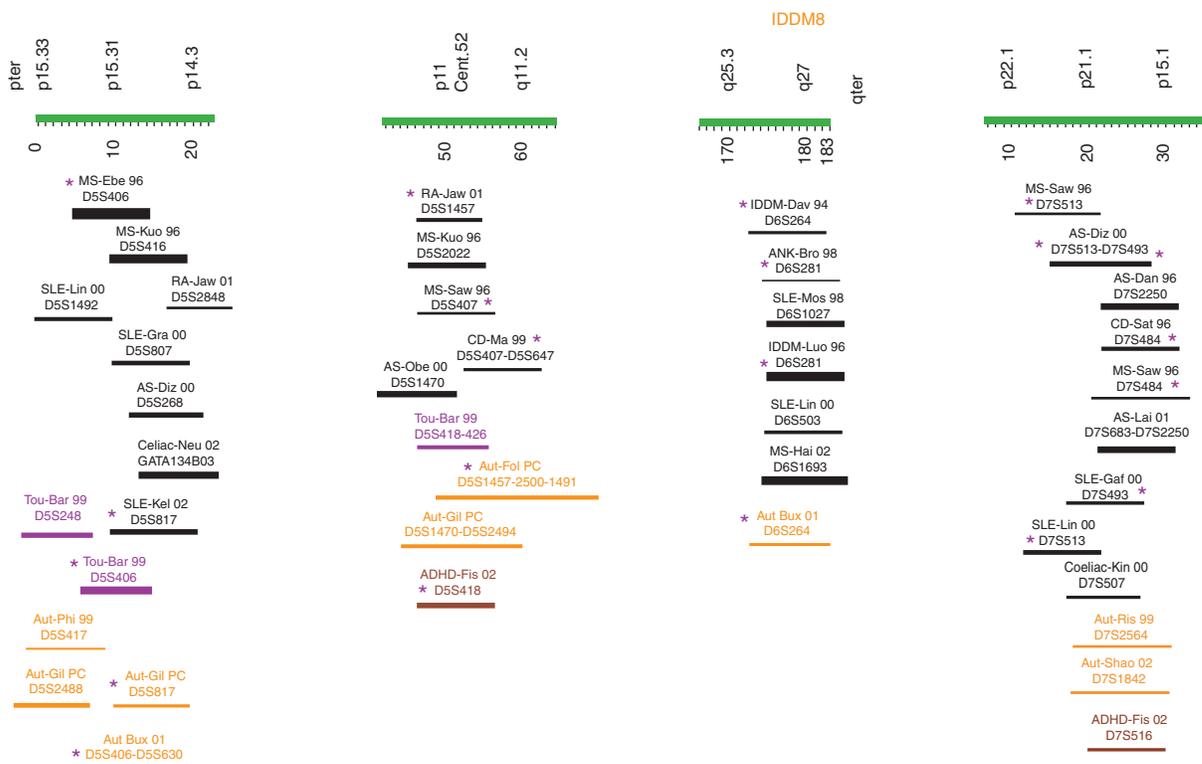
1b

5p15-p14

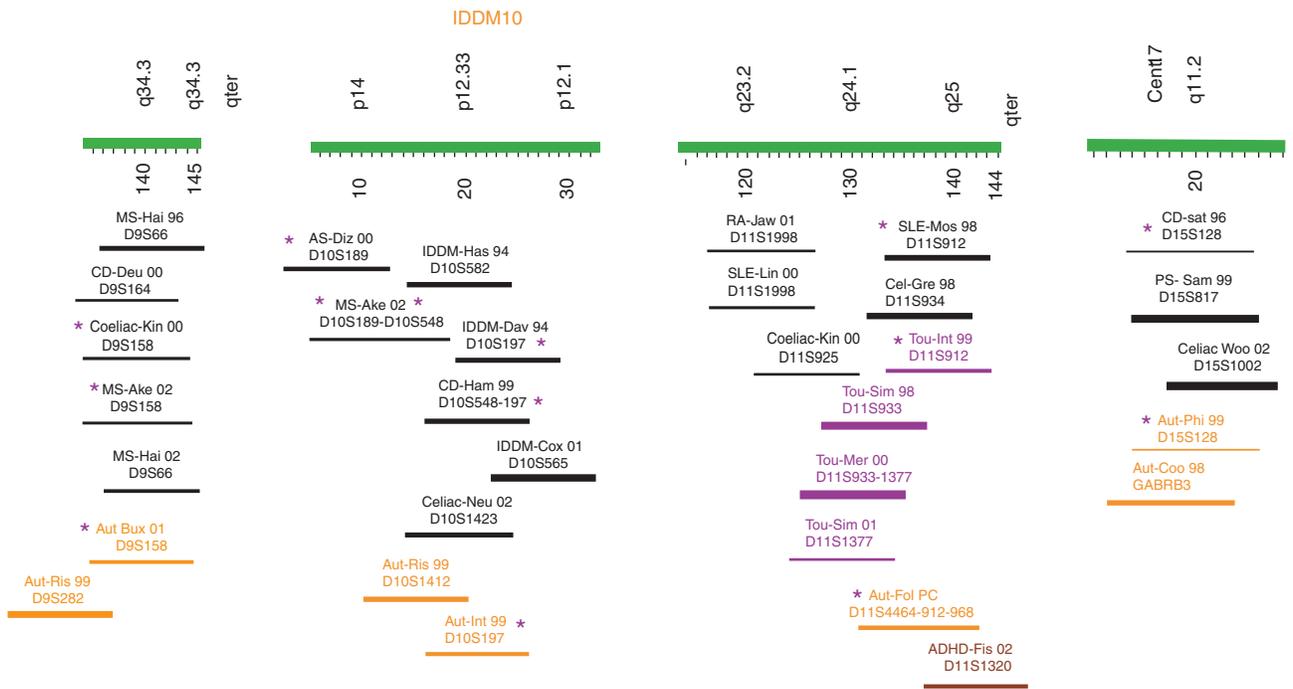
5p11-q11

6q25-q27

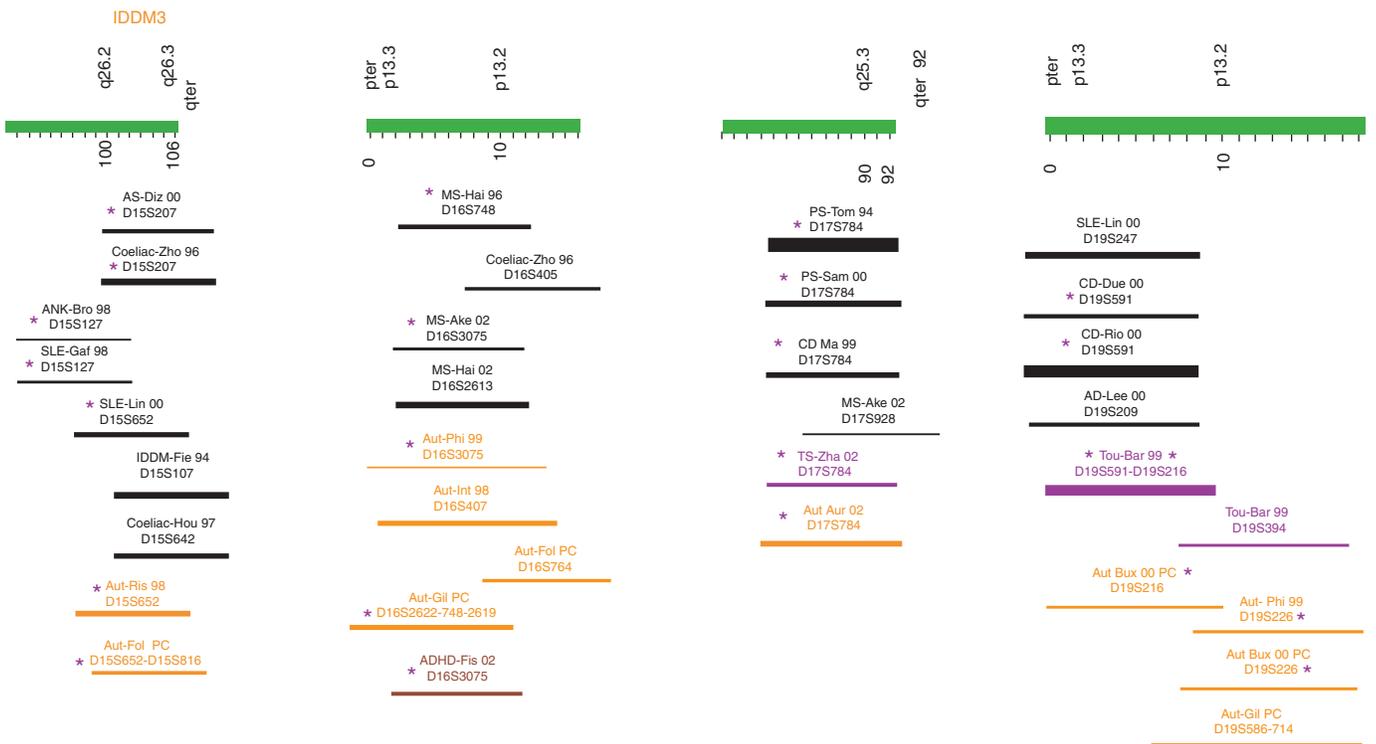
7p22-p15



1c 9q34 10p14 11q24-q25 15cen-q12



1d 15q26 16p13 17q25-ter 19p13



Supplemental Information

Read Me:

The Supplemental material provided is for review purposes.
It is the authors intent that:

Supplement 1 will take a take the final form
of individual web based images as found at this web address:
<http://www.grc.nia.nih.gov/branches/rrb/dna/atmap.htm>

Supplement 2 will take the final form of a searchable web based database as found here:
<https://www.quickbase.com/db/8jp3dz49>

Supplement 3 will also take the form of a web based searchable database as found here:
<https://www.quickbase.com/db/8qsiujvy>

Kevin G. Becker Ph.D.
Head, DNA Array Unit
Room 207
TRIAD Technology Center
333 Cassell Drive
National Institute on Aging
National Institutes of Health
Baltimore, MD 21224
TEL: 410-558-8360
FAX: 410-558-8236
beckerk@grc.nia.nih.gov
<http://www.grc.nia.nih.gov/branches/rrb/dna/dna.htm>