Molecular Genetics of Autism

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Autism is a complex, behaviorally defined, disorder characterized by impairments in 3 behavioral domains: 1) social interaction; 2) language, communication, and imaginative play; and 3) range of interests and activities. Aetiological research shows autism like a syndrome with multiple nongenetic and genetic causes. Epigenetic factors and exposure to environmental modifiers may contribute to variable expression of autism-related traits. The identity and number of genes involved remain unknown. Several studies began to focus primarily on uncovering the genetic mechanisms involved. The identification of chromosomal abnormalities and Mendelian syndromes among individuals with autism, in conjunction with data from genome screens and candidate-gene studies, has helped to refine the view of the complex genetics that underlies autism spectrum conditions. Future clinically useful insights and potential medications depend on identifying these genes and elucidating the influences of their products on brain development and physiology. **(Int. J. Ch. Neuropsychiatry, 2005, 2(2): 103-109)**

Historical Background

Autistic disorder is a behavioural syndrome beginning before the age of 3 years and lasting over the whole lifetime. It is characterized by impairment in communication, impaired social interactions, and repetitive interests and behaviour. In the first report describing autism, Kanner[1] postulated that it was an inborn disorder, yet for many years little attention was paid to possible genetic factors because it was so uncommon to have two autistic children in the same family (some two per cent of cases in the reports reviewed by Smalley et al.[2]). Early chromosome studies also failed to reveal any abnormalities. However, even a two per cent rate of autism in siblings means a huge increase in relative risk compared with the general population. The first systematic, general population based twin study, by Folstein & Rutter[3], provided evidence of a strong genetic component. Furthermore, discovery of the fragile X anomaly led to investigations with findings suggesting that a substantial minority of autistic individuals had this anomaly[4]. This led to major research interest in the association, as well as to a renewal of interest in other chromosome anomalies that might be associated with autism[5]. Clinical investigations brought growing awareness that some children with autism were affected by single gene medical disorders showing a Mendelian pattern of inheritance[6,7].

Mechanism of Inheritance

Autism is aetiologically heterogeneous. Known medical conditions are implicated in perhaps 20–25 per cent of cases, the strongest associations being with tuberous sclerosis and the fragile X syndrome. Family and twin studies show evidence of a substantial genetic predisposition in most idiopathic cases.

The frequency of autism is much higher in the siblings of children with autism (two to six per cent) than in the general population, but the strongest evidence for the heritability of autism comes from twin studies. Monozygotic twins have been shown to have a higher concordance rate than dizygotic twins in several studies.

In a review by Smalley et al.[2], pooled estimates were 61 per cent concordance rate for monozygotic (MZ) twins versus nine per cent for dizygotic (DZ) twins, giving a DZ/MZ ratio of 0.14. In a 1989 twin study by Steffenburg et al.[8],

the concordance rate in MZ twins was 91 per cent, versus 0 per cent in DZ twins. Bailey et al.[9], reported that 60 per cent of MZ twins were concordant for autism, versus 0 per cent of DZ twins. When a broad definition of the autistic spectrum was used, there was a 92 per cent concordance rate for MZ twins compared with only 10 per cent for DZ twins, giving a DZ/MZ ratio of 0.11. This suggested a heritability of autism of 91–93 per cent. The fact that the concordance rate does not reach 100 per cent in monozygotic twins is in favour of some participation by environment factors. This interaction of genetic factors and the environment corresponds to most complex human diseases.

The genetic studies indicate that autism is a genetically heterogeneous polygenic disorder caused by the additive and epistatic effect of many different genes, each with a small effect and with some showing maternal or paternal imprinting, plus a modest environmental component. While many dozens of genes may contribute to the autism phenotype, a given individual may only need to inherit a subset of these to develop autism.

The 4:1 male to female ratio for autism suggest that affected females may require a higher degree of genetic loading to develop the disorder than males. It is not known why there is a preponderance of males with autism. It is possible that one of many susceptibility genes is present on the X chromosome. This could result in a complex X linked pattern of inheritance in which heterozygous females with a susceptibility variant are at less risk than homozygous males with a susceptibility variant that is one of many genes related to autism risk. Another intriguing possibility is that there may be a gene on the X chromosome that is only expressed if inherited from the father. This would place all males at higher risk when other susceptibility factors are present.

Although altered secretion of the androgens is not a common feature of autism[10], it has been suggested that autism may arise as the result of exposure to high concentrations of prenatal testosterone.

Chromosome Abnormalities in Autism

There are several approaches that can be used in searching for genes involved in complex diseases. One of these is to find de novo chromosomal aberrations in patients and to suppose that such aberrations -corresponding to translocations or duplications-deletions-disrupt genes or regulating regions involved in the aetiology of the disease. The frequency of such anomalies in the autistic population can be estimated to be about three per cent, but unfortunately many chromosomes have been found to be abnormal. The most common specific genetic causes of autism yet identified are abnormalities of the 15q11–q13 region. Deletions of this region lead to either Angelman syndrome or Prader–Willi syndrome, depending on whether the deletion is from the maternal chromosome (Angelman syndrome) or the paternal chromosome (Prader-Willi syndrome). Both of these disorders are associated with syndromal characteristics that should lead to their identification. Both of them may be caused by having two otherwise normal chromosomes (uniparental disomy). If a patient gets two otherwise normal chromosomes from their father, they are 'missing' their mother's 15q11–q13 region (as well as the whole chromosome) and have Angelman syndrome. Angelman syndrome has been associated with autism.

Individual Candidate Genes

Various genome-wide linkage studies have been published. As is typical of the use of linkage analysis for traits that are polygenic with many genes involved, some of the results were in agreement while most were not. Most of the agreement concerned signals on chromosome 7q,on 2q, 16p (Fig. 1)[11]. The following is a review of some of the more likely candidate genes and preliminary results with some association studies.

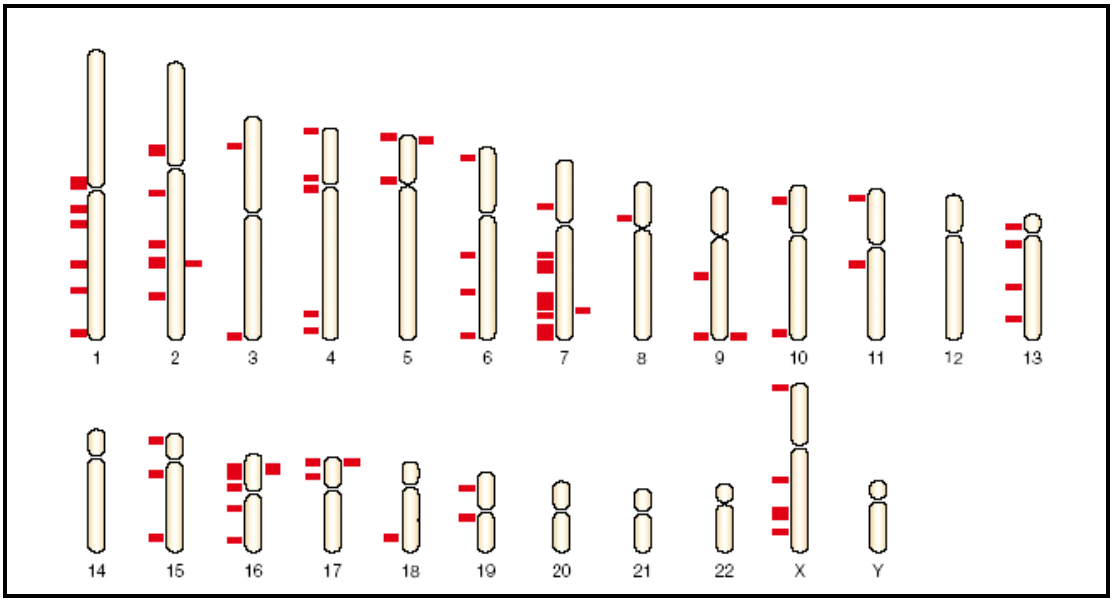

Fig. (1): Genome screen for loci that predispose to autism spectrum disorders. The most consistent signals are on chromosome 7q,2q and 16p. (From Folstein & Rosen-Sheidley, 2001.)

Chromosome 7q

The most consistent linkage finding has been the presence of a signal at the chromosome 7q31–q35 area. The 7q31 locus may have a general influence on specific language impairment (SLI). Fisher et al.[12], identified a locus on 7q31 that segregated with a severe speech and language disorder (SPCH1) in a large three generation pedigree. The trait segregated as autosomal dominant. There is considerable evidence for a gene on chromosome 7q31 that plays a role in the development of autism. Therefore SLI and autism are likely to share one or more genes in common.

Several groups have been looking closely at 7q22–31 because the region has been implicated in various genome screens. Reelin gene is expressed in this region and it has been identified in an autism association study. Moreover Hong et al.[13], identified splicing mutations in Reelin gene that cause autosomal recessive lissencephaly—an abnormality of neuron migration.

Chromosome 2

Using a sample of autism-affected relative pairs restricted to those with a delayed onset (<36 months) of phrase speech, Buxbaum et al.[14], identified a locus on chromosome 2q with a LOD score of 2.99. The chromosome 2 result is the highest multipoint maximum LOD score generated to date in an autism genome screen; it is supported by reported nominal linkage of 0.64 by Philippe et al.[15] and by IMGSAC[16].

Defects in neurotransmitters

Defects in serotonin, dopamine, norepinephrine glutamate/NMDA, GABA, and opioids have all been described in autism. Defects in the amygdala, especially in relation to problems with social interaction, have been proposed in autism, and some neurotransmitters involved in autism are well represented in this structure.

Serotonin

Serotonin plays a role in many of the behaviours associated with autism, including obsessions and compulsion, mood disorders, aggression, anxiety, impulsivity, and disorders of sleep and social interaction. One of the most highly replicated biological findings is the presence of a raised level of platelet serotonin in approximately one third of autistic subjects. Using ^{11}C-methyl-L-tryptophan and positron emission tomography, Chugani et al.[17], showed that in normal children, serotonin synthesizing capacity was more than 200 per cent that of adult levels up to the age of 5, and then declined. The decline was earlier in girls than in boys. By contrast, in autistic children serotonin synthesizing capacity increased gradually between the ages of 2 and 15 and then declined towards adult levels, and there were no sex differences. They concluded that humans undergo a period of high brain serotonin synthesizing capacity during childhood and that this developmental process is disrupted in autistic children. These results suggest that serotonin genes, and especially the serotonin transporter genes, are important candidate genes in autism.

Adenosine deaminase (ADA)

ADA plays a role in purine metabolism, immune responses, and peptidase activity. Deficiency of ADA activity gives rise to severe combined immunodeficiency (SCID). In the normal population, two alleles have been observed: ADA1 and ADA2. The second allele is associated with a decrease of 20 per cent in enzymatic activity and is the less common allele in the normal healthy European population, with a frequency of approximately six per cent. Recently two groups[18,19,20] have independently reported an association at the population level between the common adenosine deaminase biochemical polymorphism (ADA1) and autism. This result suggests that there could be linkage disequilibrium between ADA1 polymorphism and a causative polymorphism located in its vicinity. To assess whether the ADA association actually encompasses the ADA1 biochemical polymorphism, our group examined three ADA polymerase chain reaction polymorphisms in relation to autism, using pairwise measures of linkage disequilibrium in a sample of 48 autistic Italian children. Our results support the hypothesis that inside the ADA structural gene only the biochemical polymorphism (ADA1) confers susceptibility to autism. In view of the role of ADA in the normal development and function of both the nervous system and the immune system, the observed association of the less active ADA1 allele with autism justifies a specific function of the ADA gene in the pathology of autism. These investigations may benefit from biochemical explorations of purine metabolism in autistic patients.

Tuberous sclerosis

Tuberous sclerosis complex (TSC) is a rare dominant genetic disease caused by mutations of two functionally related genes, TSC1 and TSC2. Autistic disorder is common in TSC[21]. Different studies have shown that three to four per cent of autistic subjects may have TSC. Conversely, studies of TSC patients have identified much higher rates of autism, ranging from 17 per cent to as high as 61 per cent. Curatolo and co-workers[22] identified autism in six of 23 patients (26 per cent) with TSC. On the other hand, among individuals with autistic disorder the rate of TSC ranges from one per cent to three per cent, which is an increase of about 120-fold compared with the general population. This makes TSC the most common monogenic disease encountered in autistic patients. Several screens for autistic disorder have recently been completed.[23-28] Besides seven other chromosomes (1, 2, 4, 7, 10, 19, 22), a region on chromosome 16p between the markers D16S418 (16p13.2) and D16S3114 (16p13.3) shows a significant linkage to autistic disorder, with an maximum LOD score of 1.51. Philippe et al.[15], carried out a genome-wide screen with 264 microsatellites markers in 51 multiplex families, using non-parametric linkage methods. Eleven regions gave nominal P values of 0.05 or lower. One of these regions overlapped with regions on chromosomes 16p identified by the first genome-wide scan of autism.

Besides the linkage analysis in multiplex families, the existence of a susceptibility locus for autistic disorder on 16p13 is supported by other observations. Hebebrand et al.[29], reported the case of a patient with autistic disorder and a partial duplication of 16p13, and another case of autism associated with a microduplication of 16p13 has been found by Philippe et al.[15]. Moreover, autism occurs in a number of patients with tuberous sclerosis, and about half the cases of TSC are caused by mutations within the TSC2 gene mapping on chromosome 16p13 [30].

Comorbidity of autism with epilepsy, attention deficit hyperactivity disorder, mental retardation, and Tourette syndrome confirms the involvement of this region. In fact, each of these disorders had susceptibility genes in the 16p region [29, 31,32].

Our recent study[33] compared polymorphism allele frequency distributions between affected probands and unaffected control subjects for D16S291 and D16S407. We described a marginal association between the marker D16S502 (mapping 16p13.2) and autism. D16S502 polymorphism may be in linkage disequilibrium with an unidentified functional gene. Our result is the first obtained in an Italian population using an association study and seems to be in line with the relation between the 16p13 region and autism previously identified using genome screening and linkage analysis.

Future Directions

The linkage findings are consistent with a great deal of genetic heterogeneity in a true polygenic trait, with most of the genes accounting for less than three per cent of the variance. Attempts to identify the causative genes in autism using linkage techniques, such as sibling pair analysis, have been largely unsuccessful. Case–control association studies are likely to be the single most powerful methods of identifying the genes involved in autism, but parent–child trios will be necessary to test for preferential transmission from the maternal or paternal side.

Because of the great genetic heterogeneity of autism, 100 different gene variants may be involved in all autistic subjects. However, of these only 10 to 20 may be causative in a given individual. Techniques that examine genes one at a time are unlikely to identify the genes involved. With the current ability to test to many genes simultaneously in a single tube or array, autistic subjects could easily be screened for the entire set of identified relevant genes. This would show which genes and which combinations of genes are involved in a given subject. This in turn could allow treatment to be tailored for each individual, based on the genetic profile.

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