Advances in Understanding Causes of Autism and Effective Interventions

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Abstract

Understanding the aberrant biological mechanisms that underlie autism and its spectrum disorders is an exciting challenge for researchers and clinicians because breakthroughs in our understanding of the primary causes of autism are expected to lead to new approaches for intervention, prevention or even cures. This review will highlight some recent advances in our understanding of the causes of autism and of its treatment, including:

- *How some forms of autism can actually be prevented*
- *The possibly changing prevalence of autism*
- *Problems in diagnosing autism, particularly in very young children, and how such problems can complicate the interpretation of research results*
- *The many different types of autistic disorders and "double syndromes" that are associated with autistic features*
- *Different biological abnormalities in autism including abnormalities of neurotransmitters, metabolism, the immune system, and brain structure and function*
- *Suspected environmental risk factors for autism*
- *Genetic risk factors for autism*
- *The importance of animal models for autism*
- *Therapeutic approaches in autism including medical, intensive early behavioural, educational, and "alternative" interventions*

The need for multi-disciplinary teams for clinical assessment and intervention and the plights of individuals with severe autism, especially of older children, adolescents, and adults, are stressed.

Autism was first described in detail in 1943 by Leo Kanner after observing similar behaviour patterns in 11 children. He further noticed a common "extreme aloneness from the beginning of life and an anxiously obsessive desire for the preservation of sameness" (Kanner, 1943, p. 217). He referred to his children as being autistic, a term

coined in 1911 by Eugen Bleuler who used it to refer to a narrowing of relationships to people and to the outside world, a narrowing so extreme that it seemed to exclude everything except the person's own self. Hans Asperger made similar discoveries at about the same time, but the patients he identified all had speech (Fombonne & Tidmarsh, 2003). Thus, the term Asperger syndrome is often applied to higher functioning people with autism who have speech (see also Frith,1989).

There have been several controversies regarding the cause of autism over the years. For example, the main cause was assumed by some to be bad parenting. Today, however, there is general agreement that the symptoms of autism, with the exception of those of abandoned children, are a behavioural response by young children to an organic disease affecting their brains. In fact, it is now generally understood that autism is a complex developmental syndrome representing a heterogeneous group of disorders with similar symptoms, but with different biological etiologies. Realizing that autism does not have a single cause has been important for enhancing the understanding of its etiologies, prevention and treatment.

There have been many significant advances in scientific research with respect to understanding the multi-causal nature of autism. One of the most encouraging developments is that some forms of autism appear to have causes that can be prevented. There is evidence, for example, that autism is strongly associated with congenital rubella infection (Chess, 1977; Trottier, Srivastava & Walter, 1999). Since young women can now be immunized against rubella before they become pregnant, such immunization should prevent "rubella autism." Another example of a preventable cause of autism is phenylketonuria (PKU), a disorder associated with features of autism in which the amino acid phenylalanine cannot be properly metabolized (Lowe, Tanaka, Seashore, Young & Cohen, 1980). Fortunately, PKU is now largely a disorder of the past, at least in developed countries. Infant screening programs can now detect PKU during the neonatal period and early childhood. With very early diagnosis, PKU can be controlled by therapeutic diet, which ensures a low intake of phenylalanine in order to prevent further development of the disorder.

Thus, there now is growing evidence that many factors — genetic, environmental, metabolic and immunological — are involved in autism. Identifying the primary factors that result in autism is important because such knowledge will lead to better treatments, prevention, or even cures. In this paper, we review recent research advances in the field of autism, including searches for primary causes and for effective forms of intervention.

Prevalence

Curiously, autism affects boys four times more frequently than girls. The prevalence of autism is often reported to be 2-5 in 10,000 (Fombonne, 1996; Lord, Rutter & Le Couteur, 1994). This figure may be too low, as some studies indicate that the number

of children who display autistic and autistic-like conditions, as defined under the autism spectrum disorders (see Figure 1) is increasing (Bax, 1994). Some recent studies have reported a prevalence rate in excess of 20 in 10,000 children (Kadesjo, Gillberg, & Hagberg, 1999; Webb, Lobo, Hervas, Scourfield & Fraser, 1997) or 4-5 in 1,000 (Gillberg & Coleman, 2000).

Figure 1: Synonyms used in the autistic spectrum "umbrella". (After Gillberg & Coleman, 2000.)

Autism spectrum disorders, autism and its spectrum disorders, autism and autistic-like conditions, autistic continuum, pervasive developmental disorders, empathy disorders

Kanner syndrome, autistic disorder, childhood autism, infantile autism, classic autism

Asperger syndrome, Asperger's disorder, autistic psychopathology, schizoid personality disorder*, high functioning autism**

Heller syndrome, disintegrative disorder, dementia infantilis

Atypical autism, autistic-like conditions, PDD-NOS

* overlapping but not synonymous with autism
** some authors regard this as synonymous with autism, others as overlapping with Asperger syndrome

There are several reasons for the wide variation in reported prevalence rates. Some differences in prevalence may be because the diagnostic system first used in the 1960s and 1970s was different from those used more recently. One study has estimated a yearly increase in prevalence between 1966 and 1997 to be almost 4%, a highly significant value (Gillberg & Coleman, 2000). Other differences may result from a better awareness of the disorder (Wing & Potter, 2002). The need for a diagnosis of autism to procure essential services not only is increasing the awareness of autism, but this may be increasing the risk of overdiagnosing or misdiagnosing this disorder. Nevertheless, we are discovering that there may be other valid reasons why the prevalence of autism is increasing. Several studies have noted that there has been a trend for children of parents who have migrated over long distances to have autism more frequently than other children (Akinsola & Fryers 1986; Gillberg & Gillberg 1996; Tanoue, Oda, Asano & Kawashima, 1988). Maternal viral infections during pregnancy (due to lack of maternal immunity to culture-specific infectious agents) and metabolic disorders triggered by environmental factors in the new country have been suggested as possible causal factors. Thus, there is a relationship between prevalence rates, as we have measured them at any one point in time, and our knowledge of the causes and our recognition of the presenting characteristics of autism. An understanding of this relationship is essential for providing effective services.

Clinical Diagnosis

Autism is now recognized as a disorder of brain development with a strong genetic basis. It is a very heterogeneous disorder, with milder forms being more common than the classic or more severe forms. Criteria for the clinical diagnosis or classification of autism are outlined in the most recent American Psychiatric Association's Diagnostic and Statistical Manual for Classification of Mental Disorders - IV Text Revision (DSM-IV TR (2000)). Similar, though not identical, criteria are outlined in the World Health Organization's ICD-10 – another manual for classification of mental disorders that is used primarily in Europe and countries other than North America.

The DSM-IV TR criteria include some degree of impairment in reciprocal social interaction, qualitative impairment of communication, and restricted, repetitive and stereotypic patterns of behaviour, interests and activities. Because of variations in symptoms, autism is often called "autistic spectrum disorder." The term autism now includes the following:

- · Autistic disorder
- · Asperger syndrome
- · Childhood disintegrative disorder
- · Rett syndrome
- · Other autistic-like conditions (atypical autism or pervasive developmental disorder not otherwise specified (PDD-NOS))

It should be stressed that the DSM-IV TR system for "diagnosis" of autism does not address the primary cause(s) of autism. It is a classification system.

Although the DSM-IV TR provides criteria for the classification of autism, this disorder sometimes is mistaken for other developmental disabilities. Misdiagnosis of autism arises because the DSM does not provide clinicians with guidelines on how to perform the initial screenings or on which tools to use to measure behaviour. Furthermore, because the criteria include features that are deficient or absent, DSM criteria are difficult to apply to children below the age of 3 years who may not yet have developed the skills that they may acquire later.

For this reason, alternative tools are currently being developed to screen for autistic disorders, particularly in younger children (Szatmari, 2000). Some of these are more effective than others. Unfortunately, the use of non-standardized approaches for classification of autism, the failure to distinguish "autism" from other known developmental disabilities, and a tendency not to include the most severely affected individuals in research (Charmin, 1994), is causing confusion and contributing to the generation of research results that cannot be duplicated. At present, researchers are attempting to identify genetic and biochemical markers that will help with a definite

diagnosis of autism in very young children, because children with such problems should be identified as early as possible for intensive behavioural and educational intervention (see Szatmari, 2000, for a review of this topic). A more complete list of disorders that sometimes are included in the umbrella term "autistic spectrum" is given in Figure 1.

Many clinicians now believe that the causes and expression of autism are different for each child, but that there is a common pathway in the brain that results in autistic behaviours. Disorders are sometimes mistaken for autism because they have a known primary cause but are associated with autistic features. These are sometimes referred to as the "double syndromes." Some studies have reported that up to 15-30% of people diagnosed with autism actually have one of the double syndromes (Barton & Volkmar, 1998). A list of the so-called "double-syndromes" is given in Figure 2. This figure emphasizes how intertwined the features of autism can be with many different disorders.

Researchers in the field of autism are becoming increasingly aware that there are different subtypes of autism that are characterized by distinctive neurocognitive and neurobehavioural profiles (Tager-Flusbert & Joseph, 2003). For example, recognition that Rett syndrome usually affects only girls was a key factor aiding the recent discovery of the genetic mutations that cause this disorder.

The Biology of Autism

A great deal of research and funding has been devoted to understanding the cause of autism. Scientific studies presently are focussing on identifying neurotransmitter abnormalities, metabolic, genetic and environmental factors, involvement of the immune system, and structural and functional changes in the brain.

Neurotransmitter abnormalities

Some studies are characterizing involvement of the neurotransmitter serotonin in autism (Chugani, 2002; Piven et al., 1991; Veenstra-Vander Weele, Anderson & Cook, 2000) and genetic factors affecting serotonin metabolism (Williams et al., 2003). This work is contributing evidence that, theoretically, serotonin may have a special relevance to autism and other developmental disorders because of its involvement in neurogenesis – the formation of new neurons in the brain (Azmitia, Frankfurt, Davila, Whitaker-Azmitia & Zhou, 1990).

Serotonin is known to act as a trophic, or differentiating factor, in the developing brain and later as a neurotransmitter in the child and adult. Serotonin levels are higher in children and begin to decline during puberty. Many persons with autism have elevated levels of serotonin in their blood platelets, suggesting that people with

autism might have a defect in the gene that produces the serotonin transporter – the protein that removes serotonin from the space between two nerve cells. Despite this, the observation that selective serotonin re-uptake inhibitor (SSRI) drugs have improved specific symptoms in up to half the patients with autism, favours the likelihood of low serotonin in the brain in some patients with autism (McDougle, Naylor, Cohen, Volkmar, Heninger & Price, 1996). Supporting the hypothesis that abnormalities of serotonin metabolism are frequent in autism is the finding that depletion of tryptophan (a precursor of serotonin) in the diet worsens behaviour in a substantial fraction of autistic children tested (McDougle, Naylor, Cohen, Aghajanian, Heninger & Price, 1996). Complicating the picture is our knowledge that, even if there are abnormal levels of serotonin in the brain, these are not exclusive to autism, but can occur in many other diseases such as hyperthyroidism, migraine, and asthma, to name a few (Coleman & Hur, 1973).

Figure 2: Known disorders or conditions masking as "autism". (After Gillberg & Coleman, 2000.)

"Diseases" masking as autism
 Infantile Autistic Bipolar Disorder (IABD), The Zappella Dysmaturational
 Subgroup with Familial Tics, Purine Autism, Autism/Steatorrhea Syndrome,
 Almost Autism: Childhood Disintegrative Disorder, Heller Dementia

Genetic syndromes with autistic features
 Angelman Syndrome, Anorexia Nervosa, CHARGE Association, Cohen
 Syndrome, De Lange Syndrome, Down Syndrome, Ehlers-Danlos Syndrome,
 Fragile X Syndrome, Goldenhar Syndrome, Joubert Syndrome, Kleine-Levin
 Syndrome, Lujan-Fryns Syndrome, Moebius Syndrome, Mucopolysaccharidosis,
 Neurofibromatosis 1, Noonan Syndrome, Peroxisomal Disorders, Phenylketonuria,
 The Rett Complex, Smith-Magenis Syndrome, Smith-Lemli-Opitz Syndrome,
 Sotos Syndrome, Steinert's Myotonic Dystrophy, Tourette Syndrome, Tuberous
 Sclerosis Complex, Unilateral Cerebellar Hypoplasia Syndrome, Velocardiofacial
 Syndrome, Williams Syndrome

Other conditions with autistic features
 Endocrine Syndromes:
  · Hypothyroidism, Pituitary Deficiency, Congenital Adrenal Hyperplasia

 Infectious Syndromes:
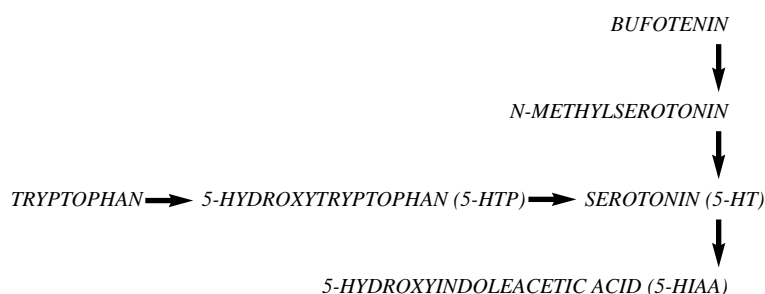 · Rubella, Herpes Simplex Virus, Encephalitis, Cytomegalovirus

 Toxic Syndromes:
  · Fetal Alcohol Syndrome, Fetal Cocaine, Exposure, Fetal Valproate Exposure,
   Lead Poisoning, Thalidomide Embryopathy

 Syndromes with multiple etiologies:
  · Blindness/Visual problems, Deafness/Auditory problems, Cerebral Palsy

Interestingly, there is recent evidence that a very high percentage of serotonin in the body (>85%) is actually localized in cells in the gastrointestinal tract, a finding that might revolutionalize our previous notion that disorders of serotonin are primarily metabolic disturbances of the brain (Weihe & Eiden, 2000). Other studies are investigating abnormalities of epinephrine, norepinephrine, the brain opioid system, and changes in oxytocin transmission in autism (Kidd, 2002a). The metabolic pathway of serotonin is given in Figure 3.

Figure 3: The metabolic pathway of serotonin (5-hydroxytryptamine, 5-HT). (After Gillberg & Coleman, 2000.)

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                                              BUFOTENIN
                                                  |
                                                  v
                                          N-METHYLSEROTONIN
                                                  |
                                                  v
TRYPTOPHAN ──► 5-HYDROXYTRYPTOPHAN (5-HTP) ──► SEROTONIN (5-HT)
                                                  |
                                                  v
                                5-HYDROXYINDOLEACETIC ACID (5-HIAA)
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Metabolic abnormalities

Biochemical factors. Though much of the research focus in autism has been on serotonin levels, the involvement of other biochemical factors has been studied. These include amino acids and organic acids, Krebs cycle analogues, melatonin, cyclic AMP, gangliosides, endorphins, lactate/pyruvate, glial fibrillary acidic protein, and catecholamines. Many recent studies have set their focus in these areas in an attempt to provide further insight that might explain differences in levels in the brains of children with autism, to identify markers that will help with the very early diagnosis of autism, and to develop metabolic approaches for intervention (Kidd, 2002a).

In addition to PKU, which can be prevented by dietary intervention, two other metabolic disorders associated with autistic features may be improved by specific dietary intervention: 5'nucleotidease superactivity (Page, 2000) and Smith-Lemli-Opitz syndrome (Starck, Lovgren-Sandblom & Bjorkhem, 2002). The frequent association of lactic acidosis and carnitine deficiency with autism has suggested that mitochondrial dysfunction resulting from excessive nitric oxide production might underlie some cases of autism (Lombard, 1998). A number of other metabolic

disorders are also associated with autistic features; these include histidinemia, adenylosuccinate lyase deficiency, dihydropyrimidine dehydrogenase deficiency, and phosphoribosylpyro-phosphate synthase deficiency (Page, 2000). Abnormalities of purine metabolism are suspected in a subgroup of people with autism who have high levels of uric acid in their urine (Page & Coleman, 2000).

Magnesium. Changes in the levels of several different ions have been reported in children with autism. Of these, the most consistent report has been that of lower levels of magnesium. Lower levels of magnesium in erythrocytes have been seen in children with pediatric psychiatric symptoms compared to normal controls (Saladino & Sankar 1973). A study of 59 children with autism displayed significantly lower serum magnesium levels than age- and sex-matched controls (Coleman, Landgrebe & Landgrebe, 1976). It is known that lower levels of magnesium in humans predispose them to apathy, irritability and seizures. As a result, children with autism, especially if they also have seizures, should be tested for magnesium levels (see Grimaldi, 2002). Magnesium therapy is reported to benefit a substantial proportion of children with autism (e.g., Galland, 1991-92; Martineau, Barthelemy, Garreau & LeLord, 1985). However, the sample sizes and methodological quality of published studies reporting beneficial effects of magnesium supplementation have been questioned (Nye & Brice, 2002; Pfeiffer, Norton, Nelson & Shott, 1995). A high dietary intake of vitamin B6 is known sometimes to result in magnesium deficiency. Thus, it is particularly important to monitor magnesium levels in individuals who are taking supplementary vitamin B6.

Xenobiotic exposure. The issue of whether children with autism have been exposed to various agents that are xenobiotic (foreign) to the central nervous system has been very controversial recently (Rice & Barone, 2000). If they have, then people with autism may have a decreased ability to detoxify these agents. In one study, abnormal liver detoxification was found in 20 children with autism (Edelson & Cantor, 1998). Other studies have suggested that perhaps a combination of detoxification problems in a child and exposure to food that is toxic to that child, especially dairy and wheat products, may be the underlying factor in the unresolved concept of diet responsive autism (Alberti, Pirrone, Elia, Waring, & Romano, 1999; McFadden, 1996; Shattock & Savery, 1997; Whiteley & Shattock, 2002).

The detoxification of foreign substances often involves adding a sulfate or a glucuronide moiety to the substances to make them more soluble and to aid with clearing them from the body. It has been hypothesized that the sulfation capacity in people with autism may be deficient and account for the high body levels of xenobiotics that have been observed (Alberti et al., 1999; Waring, Ngong, Klovrza, Green & Sharp, 1996). Waring and colleagues showed that the children they studied had reduced levels of excretion of sulfate conjugates, but no change in glucuronide metabolites. The enzyme p-sulfotransferase found in platelets was also reduced.

Based on these studies, Waring and colleagues suggested that although reduced sulfation of phenols might not be the cause of autism, it was definitely a candidate to make it worse. A deficiency of p-sulfotransferase may lead to build-ups of serotonin, dopamine and noradrenaline that are common in autism.

Possible xenobiotic exposure to either parents of children with autism, or to the children themselves in utero, has become a major political issue around the world. This has been due to apparent geographical clustering of cases. Studies are currently being conducted to determine the factors that contribute to these clusters. Possible explanations to date include coincidence, a viral epidemic, or toxic exposures (Gillberg & Coleman, 2000). Research is now focussing on following people who work with chemicals, and assessing the health of their offspring. This is a promising method of determining the effects of xenobiotic exposure. High levels of a substance called indolyl-3-acryloylglycine, a breakdown product of the amino acid tryptophan which is a precursor of serotonin, have been found in urine samples taken from many people with autism, of farmers suffering from "sheep dip syndrome" and of many chronically ill Gulf war veterans (Anderson et al., 2002). This observation suggests that there is some environmental or genetic factor common to all three conditions. Exposure to organophosphate-containing pesticides is being considered as a trigger.

Problems with the gut. Other substances related to opioids also have been found in about 80% of urine samples taken from people with autism (Reichelt & Knivsberg, 2003; Whiteley and Shattock, 2002). It has been suggested that these abnormal compounds are the consequence of an incomplete breakdown of proteins (wheat gluten and casein), which may be the result of a leaky gut. Thus, restriction of gluten and casein in the diet has the potential to be beneficial for some people with autism. Theoretically, such abnormal breakdown products might not only have the potential to affect the biochemistry of the brain but also to reduce motility of the gut and to result in constipation. About 50% of people with autism suffer from constipation (Dalrymple & Ruble, 1992). Severe bowel impaction paradoxically can result in diarrhoea; this should be treated as it can be life threatening.

Genetic factors in autism

It is now thought that the autistic spectrum of disorders (including autism, PPD and Asperger syndrome) are a result of underlying diseases with a strong genetic component (Bailey et al., 1995; Szatmari, 2003); Szatmari, Jones, Zwaigenbaum & MacLean, 1998) (see also Figure 1). Several family and twin studies have shown that hereditary factors do indeed appear to play a role in autism (Bailey et al., 1995; Bailey et al., 1998; Bailey, Palferman, Heavey & Le Couteur, 1998; Le Couteur et al., 1996). Limitations to genetic studies in autism have included the lack of a consistent diagnosis for autism and the "lumping" of people with different autistic phenotypes into one group. Still, evidence currently available suggests that although some forms

of autism may result from abnormalities in a single gene, other forms may result from abnormalities in two or more genes or an interaction between particular genes (or combinations of them) and environmental factors. It is now thought that up to 10-15 different genetic loci may be involved in autism (Risch et al., 1999). Jones and Szatmari (2002) have proposed that the effects of different genetic causes for autism are cumulatative but are not necessarily additive. It also is clear that there may be different modes of genetic transmission in autism as well as in the so-called "double" disorders (see Hallmayer et al., 1996).

Gillberg & Coleman (2000) reviewed the literature for chromosome aberrations in patients with autism. Their review includes a summary of aberrations found and molecular biology studies on each and every chromosome in the body, including the sex chromosomes. Many studies to date have also investigated regions of potential susceptibility in genome-wide scans for autism. When 99 families with evidence of genetic load were studied by the International Molecular Genetic Study of Autism Consortium (IMGSAC 1998), regions on chromosomes 4, 10, 22 and 2q, 7q, 16p and 19p were implicated. Other studies found overlapping results plus additional loci on 4q, 5p, 6q, 10q, 18q, and Xp (Philippe et al.,1999). At the present time, there is particular interest in genes possibly related to autism that lie on chromosomes 2, 3, 7, 15, and X (Shao et al., 2003). Detailed analyses of chromosomes from people with autism consistently are revealing structural abnormalities on chromosomes 7, 15 and X.

Investigation of involvement of the major histocompatibility complex locus on chromosome 6 has been ongoing since the discovery that a high percentage of autistic subjects have a variant form of the complement component 4B that produces no 4B protein (Warren et al., 1991). C4B interacts with immunoglobulin A (IgA) in the defense of the body against viral infections. Low levels of C4B may well play a key role in the pathogenesis of autism since C4 has been identified in developing brain neurons. There has been recent interest in the involvement of the HOX family of genes in autism, as this gene family turns on or off quite a number of other genes in early embryonic development. One study reporting that 40% of people with autism carry a mutated version of HOXA1 on chromosome 7 caused great excitement, but subsequent studies have failed to confirm this discovery, possibly because the participants being studied were not identified using the same criteria, guidelines or tools (Li et al., 2002).

Also of current interest is the involvement of the RLN and WNT2 genes on chromosome 7 in the autistic disorders (McCoy et al., 2002; Zhang et al., 2002). RLN codes for an extracellular protein guiding neuronal migration in development. WNT2 encodes secreted growth factor, like proteins that take part in growth regulation, differentiation, and tumorigenesis.

The recent discovery that mutations in a gene on the X chromosome called MECP2 (encoding methyl CpG binding protein 2) cause Rett syndrome, a variant of autism that affects girls and that is fatal in males, has triggered great excitement (Amir, Van den Veyver, Wan, Tran, Francke & Zoghbi, 1999). Most cases of Rett syndrome occur sporadically, that is, in the absence of any family history of the disorder. It is speculated that yet undiscovered abnormalities of MECP2 may underlie other forms of autism. Fragile X syndrome is the most common of the "double" disorders (Figure 2). Fragile X is caused by an unusual type of mutation – enlargement of a regulatory region of the FMR1 gene on the X chromosome called the trinucleotide repeat region. When such mutations are transmitted by females, the trinucleotide repeat region gets even larger. Instability of the trinucleotide repeat region in the FMR1 gene in fragile X syndrome has alerted geneticists to the possibility that phenomena involving "nucleotide repeat regions" in genes may underlie other autistic spectrum disorders. It is anticipated that most cases of autism will be the result of effects of more than one gene since monogenetic diseases (those in which single genes are responsible for a particular disease) are rare (Gillberg & Coleman, 2000). Genetic studies will be aided by the classification of affected individuals into distinct neurocognitive phenotypes based on language and cognitive profiles as well as on studies of the brain and its organization (Tager-Flusberg & Joseph, 2003).

The value of developing animal models for autism. The identification of genetic mutations that are causal in the autistic spectrum or "double" disorders has led to the development of mouse models for the disorders. Mice that lack a gene of interest are called "knockout" mice. Such animals are particularly useful for determining effects of particular genes on brain structure and function and for evaluating the safety and effectiveness of drugs on these systems before they are used in clinical settings. Synapses (connections between neurons in the brain) continually undergo changes in response to their experiences. Certain patterns of activity cause synaptic connections to strengthen. This is called long term potentiation or LTP. Other patterns of activity cause synaptic connections to weaken. This is called long term depression or LTD. This plasticity of synapses (i.e., the formation of new synapses, the disappearance of existing ones, or the strengthening or weakening of existing synapses) is thought to be the basis for most of our learning and memory.

Tranflaglia (2003) has hypothesized that too much LTD may play a role in autism. LTD is known to be controlled by one type of glutamate receptor called mGluR5 (metabotropic glutamate receptor, subtype 5). Glutamate is the major excitatory neurotransmitter. There is evidence that LTD is increased in the brains of the FMR1 knockout mouse – which is a model for males with fragile X syndrome, one of the double syndromes listed in Table 2 (Huber, Gallagher, Warren & Bear, 2002). Because mice that lack FMR1 do not produce the protein called FMRP, which is deficient in humans with fragile X syndrome, it has been reasoned that FMRP normally might regulate LTD by suppressing activity of the mGluR5 receptor. It is

being speculated, therefore, that without FMRP too much LTD causes behavioural problems in fragile X syndrome, possibly including autistic features. Excessive LTD resulting from lack of suppression of mGluR5 is amenable to drug treament. Some promising new compounds that block mGluR5 receptors and that could serve as small molecule therapies for excessive LTD are already in development (Kuhn et al., 2002). One drug called 2-methyl-6-(phenylethylnyl)-puridine (MPEP) has already yielded therapeutic effect in the FMR1 knockout mouse. It is not inconceivable that there are problems with LTD and metabotrophic glutamate receptors in people with autistic spectrum disorders other than fragile X. Thus, immediate future studies might examine mouse models of other forms of autism such as Rett syndrome or Angelman syndrome to see if these also exhibit increases in LTD, and if mGluR5 blockers such as MPEP will yield therapeutic effects. (See Tranfaglia, 2003, for further detail.)

Environmental factors

In addition to the factors mentioned above, exposures to various types of environmental factors are currently being investigated as risk factors for autism. These include lead and mercury poisoning, maternal alcohol consumption, drug abuse and smoking, exposure to valproic acid or thalidomide very early in pregnancy, and pre- or perinatal anoxia/asphyxia, as well as different types of in utero viral infections. One possible cause of perinatal axphyxia is hypothesized to be the practice of cutting the umbilical cord of babies immediately after delivery before they have taken their first breath (Simon & Morley, 2002). Autism may also be associated with the wrapping of the umbilical cord around a baby's neck at birth.

Abnormalities of the immune system

General abnormalities. Several abnormalities of the immune system have been described in people with autism. These include myeloperoxidase deficiency, severe combined immunodeficiency, IgA deficiencies (partial and complete), IgG subclass deficiencies, impaired antibody production, a skewing of T cell subsets, aberrant cytokine profiles as well as other impairments consistent with chronic inflammation and autoimmunity (Croonenberghs, Bosmans, Deboutte, Kenis, & Maes, 2002; Kidd, 2002a; Warren et al., 1997).

Deficiency of complement C4B. As mentioned above, a high frequency of people with autism have a deficiency of a protein called complement 4B, which may compromise their ability to ward off virus infection. This abnormality also may be one factor contributing to a number of immune problems described in people with autism (see above and Warren et al., 1991, for details).

Inflammation of the gut. One group has identified an unusual form of lymphocytic colitis associated with deposition of IgG and complement C1q deposition in a high

proportion of children with regressive autism (Torrente et al., 2002). Identifying the antigen(s) resulting in this unusual inflammatory response is considered to be very important as the antigens themselves, the inflammatory response that they provoke, as well as nutritional deficiencies and other problems that arise from gut malfunction, might possibly result in alterations of the mind, mood, memory or behaviour.

Measles-mumps-rubella (MMR) and other vaccines. There has been a tremendous amount of publicity surrounding the practice of innoculating very young babies with the measles-mumps-rubella (MMR) vaccine, because of concern that this may in some way be predisposing to autism in a very small number of babies (see Thrower, 2002, for further detail). This vaccine is having an enormous beneficial effect worldwide in preventing morbidity and mortality associated with measles, mumps and rubella which are very serious childhood diseases. Many large epidemiological studies have failed to provide convincing evidence of a relationship between the MMR vaccine and autism. However, some biomedical studies are suggesting a direct relationship between the MMR vaccine and autism, although these are presently controversial because findings have not been uniformly positive. For example, measles virus consistent with that in the MMR vaccine has been found in the peripheral mononuclear cells of some children who showed developmental regression and gastrointestinal symptoms soon after receiving the MMR vaccine (Kawashima, Mori, Kashiwagi, Takekuma, Hoshika & Wakefield, 2001). Another study has reported the presence of an unusual antibody to measles virus in the serum of children with autism who had been innoculated with the MMR vaccine but not in the serum of control children (Singh, Lin, Newell & Nelson, 2002). An observation suggesting that people with autism do not respond in the usual way to vaccination is the finding that a high percentage of one group of children with autism who had been innoculated against rubella did not mount a typical immune response when re-challenged (Stubbs, 1976). Another question that has been raised about the MMR vaccine is whether women should be innoculated immediately post-partum as there may be transfer of the antigen to their baby through nursing. There are concerns that some cases of autism might be associated with the inadvertent immunization of women early in pregnancy with MMR or HBV vaccines (Yazbak & Diodati, 2002).

Brain changes in autism

A challenging objective in the fields of neurophysiology and neuropathology has been trying to delineate the brain features at the gross, microscopic, and functional levels that are common to all patients with autism, as these might explain their autistic behaviour patterns. A number of new imaging technologies are aiding our understanding of abnormalities of brain structure and function (Rumsey & Ernst, 2000). These include:

· Magnetic resonance imaging (MRI). This procedure uses a magnetic field and strong pulses of radiowaves to induce protons in the nuclei of various elements to emit characteristic radio signals that are detected by a scanner and translated into an image by a computer. Structural MRI looks at brain anatomy. A more recent variation called functional MRI (fMRI) reveals regions of the brain that are activated when participants engage in a brain-stimulating task. The cerebellum may play a role in multiple functional domains including cognitive, affective and sensory as well as motor. Autopsy examination of cerebella from people with autism have revealed anatomical abnormalities of this brain region in over 90% of cases. FMRI is now being used to examine the relationship between such pathology and cognitive and motor functions within the cerebellum of people with autism (Allen & Courchesne, 2003).

· Magnetic resonance spectroscopy (MRS). This identifies and quantifies various brain chemicals via their characteristic patterns of radio signals that are emitted. One current application of this procedure is to study the relation between choline/creatine ratios and the severity of autism (Sokol, Dunn, Edwards-Brown & Feinberg, 2002). Choline is the precursor of the neurotransmitter acetylcholine. Creatine is a substance that is used to store energy in cells including the brain.

· Positron emission tomography (PET). This approach involves the injection of compounds that have a determinant that can be metabolized as well as a determinant that is radioactive. In PET, an array of scanners monitor the location and density of radioactive signal in particular tissues. One application of PET is to study serotonin synthesis and its metabolism in autism (Chugani, 2002).

· Magnetoencephalography (MEG). This measures magnetic fields around electrical currents that flow through neurons near the brain surface. It is being used to study signal processing in the primary sensory cortex, which may be impaired in autism, and also the association between epilepsy and autism, as some children with autism show epilepsy-like activity in their brains, particularly when they are asleep. It is thought that such activity might disrupt the formation of neural networks in particular brain areas and result in particular autistic features and behaviours (Wheless, Simos & Butler, 2002).

· Geodesic Sensor Net. This device uses dozens of sponge-tipped electrodes moistened in a salt solution which are placed on the scalp to map the brain's electrical activity. This device fills in the gap between conventional EEG that is fast but not too sensitive and MRI that is sensitive but slow. One recent study has suggested an association of autism with impairment in face recognition early in life (Dawson, 2002).

Children are now being given training to participate in studies using these new imaging methods.

Other important brain research involves morphometric studies of the brain, detailed histopathological examination of brain tissue, and studies of gene expression in brain sections in vitro (Acosta & Pearl, 2003; Kemper & Bauman, 1998; Kidd, 2002a). Changes that researchers have found in people with autism using these approaches include:

· macroencephaly (i.e., an increase in brain size), acceleration and then deceleration in brain growth
· increased neuronal packing and decreased neuron size in the limbic system (The limbic system is sometimes referred to as the emotional brain. It consists of an interrelated group of structures involved in regulation of the emotional state.)
· decreased Purkinje cell number in the cerebellum (The cerebellum is the primary brain structure for the coordination of groups of muscles, regulation of muscle tone, degree of muscular contraction, and performance of rapid, precise and coordinated movement patterns.)

Rodier (2002) has proposed that there is converging evidence for brain stem injury in autism. Acosta and Pearl (2003, p. 149) have suggested that "abnormalities in organization of the cortical minicolumn, which represents the fundamental sub-unit of vertical cortical organization, may underlie the pathology of autism and result in altered thalamocortical connections, cortical disinhibition, and dysfunction of the arousal-modulating system of the brain".

Therapies

One of the greatest challenges in field of autism has been the development of effective treatments. A major limitation is that behaviour is affected by a number of factors, and that these need to be considered within the context of the developing child. Aside from the prevention of rubella autism and the prevention of PKU, which is associated with autistic features, there are no specific treatments currently available that can cure any group of patients with autism. Nevertheless, therapeutic interventions that are classified as medical, behavioural, educational, or dietary, help with the management of autism (see Kidd, 2002b; Knivsber, Reichelt & Nodland, 2001; Page, 2000; Volkmar, 2001). Families should be provided with information about the range of therapeutic approaches that might be considered in the treatment of autism, work with professionals in the application of these approaches, and be encouraged to participate in well-designed trials that evaluate the outcomes of such approaches. (see Elder, 2002; Grandin, 2000; Wheatley, 2002.)

Medical therapies

Medical therapies have been reported to improve some clinical dysfunctions in particular children with autism. These medical therapies can be categorized into three types. The first type focusses on treating the basic disease process itself (i.e., treating the specific site in the metabolic pathway). As already mentioned, one example involves the prevention of phenylketonuria (PKU) by monitoring the diet throughout development. In developed countries, most babies are tested for PKU within days after birth. The second type of therapy attempts to treat the symptom complexes that are associated with the autistic syndrome, though they may not be specific (i.e., sleep disorders, hyperactivity, etc.). Last, the uses of non-specific therapies include treatment of the core symptoms of the overall syndrome. Though these drugs have a specific mode of action, their actions in the brain may involve metabolic pathways that need further research (see Volkmar, 2001, for a recent review of pharmacological interventions in autism).

Drug metabolism is the major factor involved in drug clearance from the body. Genetic polymorphisms of at least 60 different types of drug metabolizing enzymes, collectively called "cytochrome P450s" (CYPs), and changes in the expression of CYPs as the result of previous exposure to drugs, account for great inter-individual differences in the effectiveness of medications, their rate of metabolism, and drug-drug interactions (Cupp & Tracy, 1998). For this reason, "evidence based methodology" rather than "experience based methodology" (i.e., choosing a drug that has been used widely though not necessarily effectively) should be the approach of choice in medical therapy that modulates levels of neurotransmitters in individuals. Evidence based medicine is a methodology for evaluating the validity of research in clinical medicine and applying the results to the care of individual patients. It is a process of problem-based learning. The process involves:

- Converting information into one or more focussed questions
- Tracking down evidence with which to answer the questions
- Critically appraising the evidence for validity and clinical usefulness
- Applying the findings to the individual case
- Evaluating the effectiveness of the treatment in the clinical application

Refer to the Centre for Evidence Based Medicine website for further information about evidence based methodology in medicine. Medications that have been used to control rage in autism include propranolol and clonidine. Risperidone at very low doses is reported to be very effective in some cases (see also Haspel, 1995; Huggins & Homatidis, 2002; Wheatley, 2002).

Early intensive behavioural intervention and education in young children with autism and plights of older individuals with severe autism

There has been a recent explosion in the development of intensive early intervention programs – in the community, at school, and in the home – for young children with autism. Ferster and De Myer (1961) were among the first to report on the effects of behavioural intervention in the treatment of autism. On the basis of this work, Lovaas and colleagues began developing and evaluating operant discrimination learning techniques and intervention packages for very young children with autism. They reported that a significant percentage of children with autism were mainstreamed after intensive, long-term therapy compared to a much lower percentage who received less intensive behaviour modification. Furthermore, the gain resulting from the intensive intervention was found to be preserved in a follow-up study conducted several years later (McEachin, Smith & Lovaas, 1993). This treatment style has been widely used, athough the research findings have been difficult to replicate (Bibby, Eikeseth, Martin, Mudford, Reeves, 2002; Mudford, Martin, Eikeseth and Bibby, 2001). Some reasons for this are: the treatment is expensive when there is intensive (20-40 hours per week) one-on-one professional supervision, and treatment is sometimes difficult to maintain when carried out in the homes of the children with autism as their families are stressed. Further, in studies to date the inclusion and exclusion criteria, and the criteria for diagnosis of autism have not been adequately described. Also, it is not clear to what extent the participants are representative of those with autistic spectrum disorders. Pressing questions are whether the nature of the intensive intervention really matters and if there are clinical indicators that might predict which children are the most likely to show improvement with this approach. A logical extension of these studies is to ask whether intensive early intervention and education might have beneficial effects for children with developmental disabilities other than autism, for example with fetal alcohol syndrome or effects. It is not clear whether intensive early intervention will benefit children with very severe autism, especially if they are non-verbal. Unfortunately, people with severe autism, especially non-verbal older children, adolescents and adults, have not been adequately represented in research studies to date (Charmin, 1994). Also, they are having great difficulty procuring the intervention and services that they need. (See the Ontario Adults with Autism Research and Support Network for additional information relevant to adults with autism.)

Alternative medical approaches

Treatment of autism with current pharmaceuticals does not benefit the primary symptoms and can have marked adverse effects. Intensive early intervention and education programs may benefit a substantial proportion of children with autism, but these approaches do not provide cures. Recent reviews by Elder (2002) and Kidd (2002b) summarize approaches for intervention in autism that may be considered

"alternative" medical approaches. It has been suggested that alternative medical approaches be considered as adjuncts to other forms of intervention in autism (Kidd, 2002b). This rather vast array of approaches can be confusing to caregivers and professionals. Although alternative approaches are receiving publicity in the media and on the Internet, they are presently considered controversial as most have not been evaluated by well-designed research methods. Some of these approaches, however (e.g., providing people deficient in magnesium and B6 or other vitamins with supplements, or eliminating substances from the diet that cannot be well-tolerated such as dairy and wheat products), may simply be considered as optimizing nutrition. Alternative medical approaches that have been described in the literature include:

- The removal of milk and other casein dairy products, wheat and other gluten sources, sugar, chocolate, preservatives, and food coloring from the diet (Alberti et al., 1999; Braffet, 1994; McFadden, 1996; Shattock & Savery, 1997; Whiteley & Shattock, 2002)
- The controlling of intestinal yeast infection, and of infection with other parasites, and using probiotic bacteria (lactobacillus) and nutrients to correct dysbiosis and decrease gut permeability (Brudnak, 2002)
- Correction of overload of toxic metals such as mercury or lead (Shannon, Woolf & Goldman, 2003)
- Correction of iron deficiency (Latif, Heinz & Cook, 2002)
- Supplementation with sulfur-sulfhydryl compounds to aid with body detoxification (Lonsdale, Shamberger & Audhya, 2002)
- Supplementation with vitamin B6 and magnesium (Galland, 1991-92; Grimaldi, 2002; Martineau et al., 1989; Pfeiffer et al., 1995)
- Supplementation with melatonin (Lord, 1998)
- Supplementation with vitamins A, B3, C, and folic acid; the minerals calcium and zinc; cod liver oil; and digestive enzymes (Megson, 2000)
- Correction of thiamine (vitamin B1) deficiency (Simon, 1990)
- Supplementation with dimethylglycine (DMG) to boost the immune system (Kern, Hiller, Cauller, Kendall, Mehta, & Dodd, 2001)
- Supplementation with secretin, an enzyme secreted by the pancreas that aids with digestion (Cohen, 1999; Horvath & Perman, 2002; Owley et al., 1999)
- Immune therapies (pentoxifyllin, intravenous immunoglobulin, transfer factor, and colostrum) (Gupta, 2000)
- Supplementation with long-chain omega-3 fatty acids to combat excessive oxidation processes (Vancassel et al., 2001).

To achieve successful "alternative" medical management, it is important that families or individuals work hand in hand with their doctors, and ideally with nutritional and environmental specialists. As in the administration of pharmacoactive agents,

alternative medical therapy also should be administered using evidence based methodology. Clinical trials in the use of alternative interventions in the autistic spectrum disorders should be encouraged so that the results and side effects might be widely disseminated.

Summary

Breakthroughs in our understanding of the primary causes of the autistic spectrum disorders are leading to new approaches for effective intervention and a quest for prevention or even cures. Some forms of autism (e.g., rubella autism and PKU) can be prevented. The prevalence of autism seems to be increasing, possibly as the result of changes in diagnostic criteria, interest in the disorder, and exposure to new infectious organisms and dietary factors. The "diagnosis" of autism, which really is a classification system, is far from ideal, and the misdiagnosis of autism is complicating therapeutic endeavours as well as the search for primary causes of autism.

Quite a number of other disorders whose primary cause is known can mask as autism (fragile X syndrome is a common example). Autism is associated with various genetic, metabolic and immune system abnormalities. Abnormalities of serotonin metabolism affect many people with autism. Serious disturbances in the function of the gut have been identified in some people with autism, possibly resulting in complications that include vitamin and mineral deficiencies and food allergies or sensitivities. Some suspected environmental risk factors for autism include lead poisoning, perinatal anoxia (lack of oxygen), and maternal alcohol consumption. There is a strong genetic involvement in autism. Notable genetic discoveries include a confirmed high frequency in people with autism of a complement C4B gene variant which produces no protein (this may underlie some of the immune deficiencies noted in autism), and the discovery of mutations in the MECP2 gene that cause Rett syndrome, a form of autism that usually only affects young girls. New imaging methods already are detecting changes in brain structure and function in people with autism.

Therapeutic approaches for autism include medical, intensive behavioural and educational intervention strategies, but these usually only help with the management of autism. Alternative medical approaches that include dietary supplements or restrictions, or immune interventions, should be considered as a prerequisite or adjunct to other therapeutic approaches. The process of evidence based medicine, which is tailored to the individual and which examines the effectiveness of the therapeutic approach, is advocated in all forms of intervention. Prospective research studies must describe participants in sufficient detail so that others may be able to duplicate these experiments. Animal models for Rett syndrome and fragile X syndrome are leading to the development of new medications that might have general therapeutic benefit.

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Endnote

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