

INTRODUCTION

Autism spectrum disorders (ASDs) represent a diverse and heterogeneous array of conditions unified by the variable presence of specific behaviors impacting social and communicative functions (*Whiteley, 2014*). Underlying the cognitive impairments there are physiological brain problems, caused by a large number of medical factors (*Fernell et al., 2013*).

The incidence of ASD has been rising at an alarming rate over the past three decades, although the prevalence seems to differ between countries (*Elsabbagh et al., 2012*).

The ketogenic diet, modified Atkins diet, and low-glycemic-index treatment have all emerged over the past decade as important therapeutic options for children with intractable epilepsy. Whereas only a decade ago the ketogenic diet was seen as an "alternative" treatment of last resort, it has become more frequently used throughout the world. Researchers are also highly interested in using diets for neurologic disorders other than epilepsy, including autism and brain tumors (*Kossoff et al., 2009a*).

The ketogenic diet (KGD) is a nutritional approach constituted by high-fat content with adequate protein amount for growth but insufficient levels of carbohydrates for metabolic needs, thus forcing the body to primarily use fat as a fuel source (*Freeman et al., 2006*).

ASDs has been associated to metabolic dysfunction and autism is a common trait of epilepsy-associated diseases. Thus, given the beneficial effects of ketogenic diet (KGD) on epilepsy and increased mitochondrial function, its use has the potential to ameliorate some of the ASDs-associated symptoms (*Napoli et al., 2014 b*).

Very-low-carbohydrate diets or ketogenic diets have been in use since the 1920s as a therapy for epilepsy and can, in some cases, completely remove the need for medication. Recent work over the last decade or so has provided evidence of the therapeutic potential of ketogenic diets in many pathological conditions (*Paoli et al., 2013*).

The gluten-free and casein-free diet (GFCF diet) is a popular dietary intervention used by families of people with ASD in the effort to lessen core symptoms of ASD and/or gastrointestinal problems like bloating, diarrhea, and discomfort that may impact behavior. Gluten is a peptide found in grains such as barley, rye, and wheat that provides elasticity to baked goods. Casein is a peptide found in all milk and milk products (*Hyman, 2013*).

GFCF diet can ameliorate core and peripheral symptoms and improve developmental outcome in some cases of ASD. Although not fully affirmative, the majority of published studies indicate statistically significant positive changes to symptom presentation following dietary intervention (*Whiteley et al., 2013*).

AIM OF THE WORK

1. To study the role of KGD in improving the core symptoms of ASD.
2. To compare the effect of KGD versus GFCF diet in treatment of ASD.

AUTISM

Definitions of autism:

Autism is a severe neurodevelopment disorder characterized by impaired language, communication and social skills as well as by repetitive and stereotypic pattern of behavior (*Brown et al., 2010*).

Epidemiology of autism:

A) Prevalence of autism:

The number of reported cases of autism has increased dramatically in the 1990s and 2000s. This increase is largely attributable to changes in diagnostic practices, referral patterns, availability of services, age at diagnosis, and public awareness (*El-baz et al., 2011*).

ASD prevalence according to Autism and Developmental Disabilities Monitoring (ADDM) 2010 surveillance was 14.7 per 1,000 (one in 68) among children aged 8 years in the United States (*Jon, 2014*).

Another study addressed prevalence of ASD among African children, focused on Arab countries. The prevalence of ASD among children with developmental disorders in Egypt was documented as 33.6% (*Seif Eldin et al., 2008*).

B) Sex Ratio:

ASD prevalence is 23.7 per 1,000 (one in 42) among boys and 5.3 per 1,000 (one in 189) among girls (*Jon, 2014*).

C) Social Class:

Most epidemiological studies have failed to reveal an association between autism and a higher socioeconomic status. Autism is clearly seen in all social classes (*Volkmar et al., 2005 a*).

D) Age:

Autism is a chronic disorder with an onset before the age of 3 years (*Volkmar et al., 2005b*).

Types of autism:

Autism is recognized now as a heterogeneous syndrome with a broad range of behavioral symptoms and severity. The spectrum of autism-related disorders is collected under the umbrella of pervasive developmental disorders (PDDs) (*American Psychiatric Association, 1994*).

The Diagnostic and Statistical Manual of Mental Disorders fourth edition (DSM-IV) stated that pervasive developmental disorders (PDDs), include:

- (1) Autistic disorder.
- (2) Asperger's syndrome
- (3) Pervasive developmental disorder-not otherwise specified (PDD-NOS)
- (4) Rett syndrome.
- (5) Childhood disintegrative disorder.

The term "PDD-NOS" is used to describe individuals who meet some, but not all, of the DSM-IV-TR (The Diagnostic and Statistical Manual of Mental Disorders fourth edition, text revision) criteria for autistic disorder (i.e. atypical autism) (*Johnson and Myers, 2007*).

The Diagnostic and Statistical Manual of Mental Disorders fifth edition (DSM-V) was published on May 18, 2013, superseding the DSM-IV-TR, which was published in 2000, as shown in table (1)

Table (1): Diagnostic criteria according to DSM-V (*American Psychiatric Association, 2013*).

DSM-IV-TR	DSM-V
Rett disorder or syndrome was in the spectrum	Rett disorder is eliminated because it is considered a genetic disease.
Named disorders: •Pervasive developmental disorder-not otherwise specified •Asperger disorder •Childhood disintegrative disorder •Autistic disorder	These disorders will be consolidated within the category of autism spectrum disorder.
Unusual sensory behaviors were not part of the criteria.	Unusual sensory behaviors will be added to the criteria.
3 Symptom categories: •Impairment in social interaction •Impairment in communication •Repetitive and restrictive behaviors	2 Symptom categories, with more criteria per category: •Deficits in social communication and social interaction •Repetitive and restrictive behaviors
DSM-IV-TR, Diagnostic and Statistical Manual of Mental Disorders (Fourth Edition, Text Revision); DSM-V, Diagnostic and Statistical Manual of Mental Disorders (Fifth Edition).	

Causes of autism:

The pathogenesis of ASD is incompletely understood. The general consensus is that ASD has a genetic etiology, which alters brain development, affecting social and communication development and leading to restricted interests and repetitive behavior (*Muhle et al., 2004*).

1. Genetic cause

The causes of autism are still unclear, although results from twin and family studies provide evidence for a strong genetic contribution, with the probability of multiple genetic loci involved (*El-baz et al., 2011*).

Evidence for the strong genetic contribution to development of ASD is derived from the following observations (*Muhle et al., 2004*):

- Unequal sex distribution, with 4:1 male predominance
- Increased prevalence in siblings of patients with ASD compared to the general population
- High concordance rate among monozygotic twins (36 to 96 percent) (*Hallmayer et al., 2011*).

Given the complexity of ASD and the diversity of clinical manifestations, it is likely that interactions between multiple genes are responsible for ASD and that exposure to environmental modifiers contributes to the variable expression (*Bacchelli and Maestrini, 2006*).

Several full genome searches for susceptibility loci in autism have been performed. Although several areas of the genome (i.e. on chromosome 7q, 1, 2, 6, 13 and 16) have been identified as regions of interest, no specific variation in a specific gene has been firmly established as a susceptibility gene for autism (*Cook, 2001*).

The link of some HLA alleles to autism indicates the possible contributing role of these alleles to autoimmunity in some autistic children. In a case-control study carried out on 100 participants including autistic children and their mothers attending the Pediatrics hospital of Ain Shams University showed a statistically significant higher frequency of HLA DR13 among autistic patients compared to controls. It also found that HLA DR 3 frequency in the autistic patients alleles is significantly less than those of controls (*El-Baz et al., 2015a*).

2- Neurobiological Causes

A) Brain growth:

Neuroimaging studies have shown loss of Purkinje cells and cerebellar atrophy triggered by glutamic acid decarboxylase antibody (GAD-Ab) or may be fibroblast growth factors, regulating cortical size and connectivity, may be responsible for the developmental alterations (*Rout and Dhossche, 2008; Vaccarino et al., 2009*).

B) The mirror neuron system theory:

Mirror neurons are those brain cells that are active not only while one is reacting but also when one is observing others in the outside world. These neurons are viewed as deficient in children with autism (*Cheng et al., 2008*).

C) Neuronal under connectivity:

These abnormalities include diffuse differences in total and regional gray and white matter volumes, sulcal and gyral anatomy, brain chemical concentrations, neural networks, brain lateralization, and cognitive processing compared to individuals without autism (*Chen et al., 2011*).

D) Abnormal neurotransmitters:

Abnormalities in monoamines, glutamate-amino hydroxybutyrate, dopaminergic nervous system and neuropeptides (*Mcdougale et al., 2005; Sun et al., 2008*).

Decreased dehydro-epiandrosterone sulfate (DHEA-S) responses suggest that autism is accompanied by a major dysequilibrium in the serotonergic system (*Croonenberghs et al., 2008*).

E) The intense world syndrome:

The autistic brain is hyper reactive with hyper plasticity of local neuronal circuits, leading to hyper perception, hyper attention, and hyper memory. This hyper functionality then turns debilitating, and the excessive neuronal processing

renders the world painfully intense, hence the “intense world syndrome” (*Markram et al., 2007*).

(F) Serotonin and autism:

Disorders of serotonin metabolism or disruption of serotonergic development can leave permanent alterations in brain function and behavior and this may be the case in autism (*Narita et al., 2002*).

(G) Cholinergic activity and autism:

Abnormality of Cholinergic activity is reported in the cerebral cortex and basal forebrain in autism. So, the intervention in autism should be based on cholinergic receptor modulation (*Perry et al., 2001*).

(H) Glutamate neurotransmitter and autism:

N-methyl-D-aspartate (NMDA) subtype of glutamate receptor, which is central to the developmental processes including neuronal migration, differentiation, and plasticity, is receiving an increasing attention in its relation to autism. In one study, no difference was found between NMDA receptor antagonist (amantadine) and a placebo on parent ratings, although clinician-rated measures of hyperactivity and inappropriate speech showed significant improvement (*McDougle, 2002*).

Neurogenetic syndromes that seem to play a causative role or are associated with ASD include:

- **Fragile X syndrome (FXS)**

Almost 60% of the children with FXS met criteria for autism (or a pervasive developmental disorder). Children with FXS and autism are at risk for impaired motor abilities (*Zingerevich et al., 2009*).

- **Neurocutaneous disorders (Tuberous sclerosis)**

Almost 17 to 60 percent of patients with tuberous sclerosis complex are also autistic; however, only 0.4 to 4 percent of patients with autism have tuberous sclerosis complex (*Numis et al., 2011*).

- **Rett syndrome**

Rett syndrome should be considered in all females who demonstrate autistic-like regression, especially if they have microcephaly, seizures, and hand-wringing stereotypies (*Gonzales et al., 2005*).

- **Smith-Lemli-Opitz syndrome**

It is an autosomal recessive disorder of cholesterol biosynthesis (*Johnson and Myers, 2007*).

3. Metallothionein (MT) dysfunction:

MT is a family of proteins that controls the copper/zinc ratio in the body. Children have abnormal copper/zinc ratio

with high body copper and low body zinc. Other functions of MT in the body include development of brain neurons, detoxification of heavy metals, maturation of the gastrointestinal tract (GI), anti-oxidation, boosting immune function and delivery of zinc to cells.. MT dysfunction could be caused by a genetic defect, or an environmental insult that disables MT (*Jepson, 2003*).

4. Endocrine factors

As autism occurs more frequently in males than in females, sex hormones may have role in the development or expression of autistic traits. Estrogens and progesterone have been reported to have neuroprotective and neuroregenerative role. The implications of this research are that if environmental or genetic factors or their interaction cause damage to the developing brain, this damage might be ameliorated by the presence of estrogen and progesterone. Thus females would be expected to show fewer sequel of neural damage than males (*Galinat, 2008*).

Also, it has been suggested that autistic patients have elevated blood androgens. In a study done to assess serum androgen levels in a group of Egyptian male autistic children and adolescents and their relation to disease severity , results showed that androgens were higher in autistic patients than in controls and increased with increased autistic severity. Thus, androgen levels should be assessed in autistic patients (*El-Baz et al., 2014 a*).

5. Immunological abnormalities:

Abnormalities of both humoral and cellular immune functions have been described in some studies of children with autism and include decreased production of immunoglobulins or B and T-cell dysfunction (*Volkmar et al., 2005b*).

Raised levels of brain-specific autoantibodies were reported in children who had autism (*Wills et al., 2007*). Folate receptor autoimmunity may represent an important factor in reduced folate transport in autism (*Ramaekers et al., 2007*).

Several studies provided strong evidence against the hypothesis that MMR vaccination causes autism. Combination of vaccines as MMR and DPT may also overstimulate children immune system that start the autistic biomedical cascade (*El-Baz et al., 2011*).

6. Mitochondrial causes:

Over the past decade, evidence has accumulated that some individuals with ASD have concomitant mitochondrial dysfunction (MD), and some have proposed a ‘mitochondrial autism’ subgroup. The prevalence of MD in ASD is 5 % (*Rossignol and Frye, 2012*).

Autism may be a disorder of fatty acid metabolism due to a possible dysfunction of mitochondrial enzymes responsible for the beta oxidation of unsaturated fatty acids in the mitochondria (*Clark and Clark, 2004*).

Brain energy metabolism of many autistic children is low due to summation of several factors including low plasma polyunsaturated fatty acids (PUFAs) and/or disturbed mitochondrial function as evidenced by decreased serum carnitine and increased plasma lactate levels (*Mostafa et al., 2005*).

7. Environmental factors:

A- Infection

It has been hypothesized that autism may be related to the existence of persistent infection such as herpes simplex, varicella zoster, and Epstein-Barr virus, that persist in the human body throughout life.. At present there is no evidence that such persistent infections are causally related to autism (*Brown et al., 2010*).

B- Intrauterine infection or inflammation:

Intrauterine rubella or cytomegalovirus (*Larsson et al., 2005*).

Studies proved elevated levels of certain cytokines in the cord bloods of infants who were later diagnosed as autistic with or without cognitive impairment (*Nelson et al., 2006*).

C- Low Birth Weight and Gestational age

Abnormal gestational age, including prematurity and postmaturity, and LBW has been associated with an increased risk of autism in some, but not all, studies (*Kolevzon et al., 2007*).

D- Hypoxic Conditions:

Several investigators have hypothesized that a perinatal conditions that indicate prolonged or acute oxygen deprivation (hypoxia) to the fetus may be a major risk factor for neuropsychiatric disturbance (*Kolevzon et al., 2007*).

E- Maternal diseases:

Maternal hypertension, albuminuria, and generalized edema, chorioaminionitis, acute intrapartum hemorrhage (*Wallace et al., 2008*).

F. Extreme male brain theory:

High maternal intrauterine testosterone levels (*James, 2008*).

G. Hyperbilirubinemia:

Infants who had hyper-bilirubinemia after birth had an almost fourfold risk for autism (*Maimburg et al., 2008*).

H. Heavy metal exposure:

A study was conducted on 100 children with autism in comparison to 100 controls to assess the exposure to mercury, lead, and aluminum as environmental risk factors. All participants were subjected to clinical evaluation and measurement of mercury, lead, and aluminum through hair analysis which reflects past exposure. Results showed that levels of mercury, lead, and aluminum in hair of the autistic