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Autism Spectrum Disorder (ASD): From Molecular Mechanism to Novel Therapeutic Approach

Hagit Friedman

Abstract

Autism spectrum disorder (ASD) is the joint name for neurodevelopmental impairments characterized by abnormal social interaction, communication difficulties, limited range of activities and areas of interest, and typical motor impairments. There is a remarkable increase in the prevalence of ASD over the past 30 years. Studies indicate that genetic, neurological, and environmental factors are involved in the emergence of ASD, and recent works describe the neuromolecular mechanism implicated in the basis of ASD. 3LT has now developed into a therapeutic procedure that is used for three main goals: to reduce inflammation, edema, and chronic orthopedic disorders; to promote healing of wounds, deeper tissues, and nerves; and to treat neurological injuries and pain. 3LT may treat neurological injuries by lowering levels of inflammation proteins and by stimulation of mitochondria to increase the production of adenosine triphosphate and neural growth factors. This review aims to discuss the current evidence for the effects and mechanisms of 3LT at the cellular level and the effects of 3LT-induced changes in brain development and function. Early and effective intervention, through the developmental time window of high ASD susceptibility, using tools that are directed to the mechanism of pathology, may minimize neurological and functional deficits.

Keywords: brain development, brain injury, ASD, autism, 3LT, low-level laser therapy, mitochondria

1. Introduction

Autism spectrum disorder (ASD) displays early in child development, during the time of human synapse formation and maturation [1], and usually results in long-term difficulties in social, communicational, emotional, adaptive, and cognitive functions [2]. The frequency of ASD occurrence continues to rise—from 1:110 in 2006 to 1:54 in 2016 [3], with at least one diagnosed coexisting neurodevelopmental disorder in most of the children [4]. Early diagnosis and treatment are very important as they may minimize neural injury and functional difficulty.

As ASD is still diagnosed only by behavioral criteria, it has been difficult to connect the numerous neurophysiologic findings to the clinical characteristics of ASD and to draw the mechanism and etiology of ASD [5]. This would allow an accurate treatment, directed to the mechanism of injury, with the best chance to make a change in the impaired developmental route.

The search for ASD brain mechanism may be reviewed from the neural circuit to the molecules and organelles involved.

In the late nineties of the twentieth century, a laboratory in Italy first documented neural activity from brain cycles, later named “Mirror Neurons” [6]. The innovation in its discovery was that it connected fields of neural control that were considered separate—motor and vision, that is, the same specific neuron cycles work both when a person does something and when he or she watches another person perform the same action, making an instant translation from visual to motor control [7]. This act of neural translation is considered the basis of the human ability to imitate, to anticipate others’ goals, and to empathize others’ pain or misery [8–11].

“Mirror Neurons” brain cycles showed altered activity in children with ASD, hinting that they are involved in the mechanism of ASD [12–14].

The scientific findings about mirror neurons and the possibility that their development may be related to the time window of temporary subcortical plate neurons (connecting thalamic and future cortical cycles) are indeed amazing [15]. But the mechanistic discussion in the level of neural cycle leaves many open questions—what may cause damaging alterations in these brain cycles? What cellular and molecular components are involved, and how can we target the therapeutic process to them?

Loss of synaptic stability and plasticity, or dysregulation of activity-dependent signaling networks that control synapse development, function, and plasticity, may cause injuries in neuronal circuits and contribute significantly to brain diseases, including ASD pathogenesis [16, 17].

Hence, alterations in synapse function, synaptic molecules, receptors, and neurotransmitters have been targets to research about the mechanism of ASD syndrome for the last 20 years. Studies showed that alterations in Glutamate receptors and enhanced GABA receptor-mediated inhibitory synaptic transmission are involved in ASD [18–20]. There may be various causes involved in psychiatric and neurologic diseases, including ASD—genetics, drug use, neurodegeneration, viral infections, and more. However, dysfunction of neuronal synaptic communication is almost always the underlying cellular mechanism. Epigenetic changes in synaptic genes encoding for synaptic adhesion molecules (neurexin, neuroligin, and N-cadherin) and for PSD proteins (i.e., Shank1, Shank3, and more) are involved in neuropsychiatric disorders including ASD, causing alterations in synaptic transmission [16, 21–25]. Studies have found that failure of the cellular machinery in pathways upstream of the synapse leads to synaptic dysfunction and neuropsychiatric characteristics. In addition, small non-coding microRNAs that repress the translation of target mRNAs seem to be important pathophysiologic mechanisms for neurologic and psychiatric diseases, and abnormal regulation of protein turnover, chromatin remodeling, and genomic imprinting may lead to synapse pathology. In some neuropsychiatric disorders, the basic neurobiological mechanisms underlying the symptoms are simple and easily solved, but the model of loss of function of a single gene or a limited number of genes is not suitable for most neuropsychiatric disorders, which are etiologically heterogeneous and complex and likely determined by the combination of variants/defects in multiple genes. For example, genome-wide association studies identified polymorphic variants in genes encoding synaptic proteins as important determinants of the risk of developing ASD [26–28].

2. Molecular mechanistic common denominator involved in ASD etiology

Multiple studies show that a mitochondrial disease or abnormality is involved in the etiology of ASD [29, 30] affecting about 80% of the children with ASD.

Mitochondria are the “cell powerplants,” being responsible for most of cell energy production. Sufficient energy is required for everyday vitality and for brain survival and function. Brain cells need a lot of energy to function. Apart from energy production, mitochondria participate in the cellular metabolic processes of iron and the balance of calcium. The mitochondria are associated with normal and abnormal cell proliferation and participate in programmed cell death. Each cell has hundreds to tens of thousands of mitochondria, depending on the role and energy consumption of that cell. Mitochondria are inherited only from the mother, through the ovum. They can develop mutations as they multiply and lack almost any repair mechanisms. Most of the proteins that make up mitochondria are encoded in the nucleus. Only 13 proteins are encoded by the circular mitochondrial genome.

As the mitochondria are inherited from the mother, hence, we do not have a “backup” from the father’s genome when mutations or damage occurs. But since the ovum contains a lot of mitochondria to start with, some may be damaged without any clinical manifestations. Mitochondrial damage may be manifested over the generations; when the grandmother had a few damaged mitochondria, the mother happened to develop from an ovum with a greater concentration of damaged mitochondria, and her son already has very few normal mitochondria. A problem is revealed in such cases. A damage to mitochondria may be caused not only by maternal inheritance, when cells divide to form the fetus, but also by a coding error called “de novo mutation” (a new mutation in fetal cells or in mitochondria), due to environmental / epigenetic influence. Hence, when a diet contains fewer carbs, there is an increase in the number of mitochondria in liver and large muscle cells.

Mitochondrial abnormalities include either decreased [29, 31, 32] or increased [33–36] mitochondrial function; depending on the cause and developmental time window, they may lead to neurodevelopmental regression [30, 37–42] and the typical comorbidities of ASD (i.e., gastrointestinal problems, seizures, tiredness, and sensory dysregulation) [30, 43, 44]. The first findings, leading to this conclusion go back to the eighties of the twentieth century [45], reconfirmed about 20 years later [46] and continue with studies that examine the biomarkers of mitochondrial dysfunction [30, 47]. Neurodevelopmental regression, as typically described for many children with ASD, may be the hallmark of a mitochondrial disorder and abnormal mitochondrial physiology in ASD [38, 39].

As mitochondrial function is highly influenced by environmental factors, these findings connect mitochondrial dysfunction in ASD with environmental hazards [29, 30].

3. Therapeutic approaches

Since ASD was first defined, numerous treatments have been employed, with partial/sporadic mechanistic justification. Most of the treatment approaches target behavioral abnormalities of children with ASD and aim to improve the social and communicational function of the patient [48–50].

The website of the American Association of Communication Clinicians describes 30 common treatment programs for children with autism, divided into seven classes; however, parents cannot be given definite treatment recommendations, because of the heterogenous characteristics of children with ASD and because many therapies have not yet been investigated in a controlled and satisfactory manner.

In November 2020, the Australian governmental CRC top organization published a 502-page document written by 12 scientists. The paper is a meta-analysis based on 58 review articles analyzing more than a thousand research articles that examined the effectiveness of 111 different autism therapy programs [51].

The authors sorted the programs into nine categories (cognitive, behavioral, educational, developmental, animal assisted, sensory-based, naturalistic, technology-based, others). The review showed that intensive behavioral programs achieved good results, but the results were focused on specific goals in which the child has been practiced; only some of the developmental plans showed improvement, mainly programs that included parental involvement; only one sensory program has achieved clear results of reducing stimulation and improving learning habits and participation in the community; music therapy helped interpersonal communication and improved mental well-being in the family; various computer applications have improved cognitive ability but not mutual communication; alternative supportive communication programs have resulted in good results in communication, motor behavior, game levels, and learning ability. The authors note that in each category, only a very small number of studies were made in a controlled and satisfactory manner, meaning that the results should be treated with caution.

Altogether, children with autism spectrum disorders can be treated in a way that will lead to functional and communicational improvement, using various therapeutic approaches. These treatment plans are tailored to the unique behavioral profile of each child and each family at each point in time throughout their life journey with autism. However, as these treatments focus on external behavioral symptoms, and not on the internal mechanism, they aim at functional improvement and not actual repair of neurological damage. Hence, according to this approach, autism is not a “curable injury” but a developmental disorder whose treatment helps patients develop functional skills, improve communication skills, and rely on their strengths despite the disorder that will always remain a part of their lives.

Should we be satisfied with the important achievements of symptom-oriented therapeutic approach, or perhaps a persistent search into mechanistic questions may lead to a mechanism-oriented therapeutic approach?

Few therapeutic approaches for mitochondrial disorders were examined in clinical studies in children with ASD. These include cofactor supplementation and ketogenic diet. Nutritional supplements aimed to support the mitochondria, redox, and folate pathways, and contained L-carnitine, coenzyme Q10, and additional factors. They improved mitochondrial function and ASD symptoms [52–57]. However, discontinuation of the supplement treatment caused worsening of the ASD behavior in children [31, 58].

Ketogenic diet has been studied for ASD, resulting in a mild-to-moderate improvement with 58% of the children who tolerated the 3-month diet [59–61]. In one out of three studies, worsening outcomes were observed. In the studies that used biomarkers to better understand the physiology of the ketogenic diet, an increase in chromium and creatine and a decrease in ornithine, acetoacetate, cesium, and N-acetylserotonin across the treatment period correlated with better outcomes [60, 61]. In addition, the ketogenic diet improved sociability and repetitive behaviors in two environmentally induced mice models of ASD [62–64]. With these results, the ketogenic diet needs more study for its use in children with ASD. In addition, the important limitation of the ketogenic diet is the child’s ability to tolerate the diet, as dietary therapies are difficult to implement with children. For other dietary treatments, outcomes are related to the ability of the family to implement the diet adequately [65], and if it is impossible for the family to apply the diet properly, the expected outcome may not be achievable and other therapeutic options may be a better choice.

4. Low-level laser therapy (3LT)

Alternative medicine has become vastly used for managing health problems and developmental injuries in the modern western world, consisting of various

approaches stemming from traditional medicine combined with modern empirical techniques [66, 67].

Acupuncture and auricular therapy have been employed all over the world for the treatment of chronic and acute medical situations [68–71], for coping with pain in elderly [72, 73] and children [74–78]. For example, it was found that acupuncture increases the secretion of the natural neuromodulator adenosine, also known as anti-inflammatory and pain relief substance [79].

Lasers (light amplification by stimulated emission of radiation) are devices that generate electromagnetic radiation, which are uniform in wavelength, phase, and polarization. Low-level laser (3 L) is a special type of laser that affects biologic systems through nonthermal means [80, 81]. Low-level laser therapy (3LT) is the application of red and NIR (near infrared) light over injuries or lesions to improve wound and soft-tissue healing, reduce inflammation, and give relief for both acute and chronic pain (analgesia) [82–84].

3LT applies a therapeutic laser for the excitation of specific acupuncture points. This technique is considered nonintrusive, safe, and painless [85] and became an important tool for the treatment of patients at risk, such as premature neonates [86–92]. For example, excitation of specific pain acupuncture points using 3LT creates a local photochemical effect [93] that causes specific changes in neuronal brain activity [94, 95], apprehended by the patient as reduction in pain severity. These changes can be measured and quantified by imaging [96, 97].

3LT has a photochemical effect, meaning that when the correct parameters are employed (intensity and location), red or NIR light reduces tissue oxidative stress and increases ATP levels [98–101]. This improves cell metabolism and reduces inflammation. In addition, 3LT was proven to increase nociceptive threshold by altering the axonal flow [102] and elevate opioid-receptor binding [103] and endorphin production [104].

In the clinic, 3LT was found to cause an immediate decrease in acute and chronic pain and an increase in function [102, 105–107]. 3LT showed promising results for myocardial infraction [108], rejuvenating mesenchymal stem cells [109], skin injuries [110–113], brain trauma, TBI [114–116], diabetic retinopathy [117], oncology [118], and more.

3LT is a technique of noninvasive stimulation of which the irradiation of specific infrared wavelengths can penetrate the body [119]. These effects produce various biological responses, such as enhancing the formation of adenosine triphosphate (ATP), deoxyribonucleic acid (DNA), and ribonucleic acid (RNA); releasing nitric oxide (NO) and cytochrome c oxidase (CCO); regulating reactive oxygen species (ROS); and altering intracellular organelle membrane activity, mainly in mitochondria, calcium flux, and stress proteins [66, 120–124]. 3LT produces a shift toward higher oxidation in the overall cell redox potential [125] and briefly increases the level of ROS [111, 126]. This change in the redox state of the mitochondria regulates several transcription factors [127]. These include redox factor-1 (Ref-1), cAMP response element (CREB), activator protein 1 (AP-1), p53, nuclear factor kappa B (NFjB), hypoxia-inducible factor (HIF-1), and HIF-like factor [127]. The activation and regulation of redox-sensitive genes and transcription factors are thought to be caused by ROS induced from 3LT [126]. In turn, both ATP levels and blood flow increase, improving oxygenation found in damaged areas of the brain [127].

5. Therapeutic potential

A wide range of seemingly unrelated disorders, such as schizophrenia, bipolar disease, dementia, Alzheimer's disease, epilepsy, migraine headaches, strokes, neuropathic pain, CP, TBI, diabetic retinopathy, Parkinson's disease, ataxia,

transient ischemic attack, cardiomyopathy, coronary artery disease, chronic fatigue syndrome, fibromyalgia, and SARS-CoV-2, have underlying pathophysiological mechanisms in common, namely reactive oxygen species (ROS) production and the accumulation of mitochondrial DNA (mtDNA) damage, resulting in mitochondrial dysfunction [114, 128–130].

3LT has been long recognized as an efficient therapeutic tool for brain injuries. Recent deciphering of the role of mitochondria in ASD etiology and in the 3LT therapeutic process gives us a great opportunity to improve mitochondria function and brain neural development, using suitable parameters of 3LT energy on specific ear and body locations.

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