# OXIDATIVE STRESS AND AUTISM: AN OVERVIEW

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**ABSTRACT**

Autism spectrum disorder incidence raised rapidly in the last three decades, thus becoming, a global major public health problem. In the attempt to find a solid aetiology for autism, numerous pathophysiological hypotheses have been postulated over the years, one of them being based on the imbalance of antioxidative and oxidative stress systems. In this paper, our aim is to shortly review the role of oxidative stress in the pathophysiology of autism spectrum disorder, focusing on changes of antioxidant enzymes activity and several biomolecules that act as indirect oxidative stress markers. Improving the antioxidant defence and decreasing the oxidative stress are two therapeutic interventions with a real future potential to reduce or treat autistic spectrum disorder children.

**Keywords:** autism, oxidative stress, antioxidant, review.

## 1. INTRODUCTION

Autism Spectrum Disorder (ASD) is a complex condition that involves challenges in social interaction, speech and nonverbal communication and restrictive and repetitive behaviours beginning in the early developmental period [1]. The incidence has increased dramatically over the past two decades [2]. ASD affects approximately 1 in 68 children in United States of America [3]. The aetiology of ASD is complex, involving genetic and environmental triggers, but the clear causes remain elusive at this moment. More than 100 genomic differences have been reported in children with ASD but only several of them have a strong association, meaning that only a small percentage of cases is affected by genetic defects [2].

Besides genetic triggers, metabolic abnormalities have been noted in individuals with ASD, especially in the redox balance and oxidative stress, immune regulation and inflammation and mitochondrial function.

The imbalance between reactive oxygen species production by pro-oxidants and the activity against reactive oxygen species by antioxidants may cause oxidative stress in autism spectrum disorder. Antioxidant enzymes (such as superoxide dismutase, glutathione peroxidase and catalase), glutathione, ceruloplasmin, transferrin, vitamin E, C and A, Mg, Zn, Se act as an antioxidant defence in individuals with ASD, protecting cells from the reactive oxidant species damage. On the other hand, endogenous pro-oxidants (nitric oxide, xanthine oxidase, homocysteine, circulating cytokine) and exogenous pro-oxidants (viral infections, retinoic acid, valproic acid, heavy metals, pathogenic bacteria, hexane, pentane) promote production of reactive oxygen species and consequently increased oxidative stress. The increased production of free radicals determine lipid peroxidation, protein and DNA oxidation and also mitochondrial damage leading to oxidative stress in autism. Genetic

factors have an influence in the free radicals synthesis, potentially leading to oxidative stress [4], [5].

## 2. OXIDATIVE STRESS IN THE BLOOD OF INDIVIDUALS AFFECTED BY AUTISM

Several studies have reported lower activities of antioxidant enzymes in children with ASD compared to healthy controls. Superoxide dismutase (SOD) is a potent protective enzyme that acts to catalyse the transformation of $O_2^-$ to $H_2O_2$.

Investigation of red cell superoxide dismutase, glutathione peroxidase (GSH-Px) and erythrocyte levels of GSH-Px in autistic children revealed lower levels of antioxidant defence enzymes compared to healthy controls [6, 7]. Also, Golse et al. (1978) measured platelet SOD and GSH-Px in autistic children, resulting in lower levels of antioxidant enzymes in affected children versus controls [8]. A lower glutathione (GSH) and redox ratio of reduced to oxidised glutathione (GSH/GSSG) are an indirect marker of impaired antioxidant defence and of an increased oxidative stress state.

Regarding these markers, individuals with ASD have a decreased level of glutathione and GSH/GSSG [9]. Oxidative stress potentially has implications in the male to female ratio of 3:1, observed in children diagnosed with autism spectrum disorder [10]. More precisely, males have significant lower GPx and SOD antioxidant activity and levels of GSH when compared to females thus male mitochondria is more vulnerable to free radical damage than female mitochondria [11]. Another implication of mitochondria and GSH are the gastrointestinal problems such as chronic constipation which is commonly found in autistic children. The gastrointestinal system is dependent of a normal GSH level in the inner mitochondrial membrane to function normally. Abnormal mitochondrial respiration function is linked to a depletion of GSH in children with ASD. This, in turn, can cause gastrointestinal problems [12].

Catalase has the role to decompose $H_2O_2$ to $H_2O$ and $O_2$, therefore protecting cells from the oxidative effects. In a study conducted on 27 autistic children and their healthy controls, catalase blood levels were significantly lower than controls [7].

Using indirect markers, such us oxidised biomolecules, a significant number of studies revealed an increased oxidative stress state in children with autism. Lipids - essential membrane components - are easily oxidised, especially the unsaturated ones. This oxidation leads to end products, such as malondialdehyde, that are used as an oxidative stress markers. Children with ASD have higher levels of serum MDA compared to their match control group, probably due to an increased oxidative stress state [7, 13]. The comparison was made also with their healthy sibling, with whom theoretically they share a genetic history. The brain and neuronal tissues have a great concentration of polyunsaturated fatty acids (PUFA), contributing to signal transmission, inflammation and cells' survival and repair. PUFA resides are correlated with the ASD severity. The hyperactivity symptoms are directly correlated with PUFA levels – high levels of hyperactivity link with lower levels of neural lipids [14]. This finding supported the idea of therapeutic intervention with supplementation of omega-3 fatty acids in ASD children. Two randomised double blind studies have reported an alleviation of hyperactivity symptoms after omega-3 supplementation [15, 16]. Unfortunately, a Cochrane meta-analysis concluded that omega-3 supplementation in autism lacks high quality evidence [17].

Ceruloplasmin is a brain protein with an important antioxidant role, transporting 95% of copper in the blood and protecting the membrane polyunsaturated lipids from oxidation. Ceruloplasmin also acts as a ferroxidase. Transferrin is another protein of

major importance, being responsible for the transport of iron to growing tissues acting thus as a growth factor. Ferrous iron is a catalysing agent, converting hydrogen peroxide to more toxic radicals. Ceruloplasmin and transferrin act as antioxidant compounds by lowering the free ferrous iron in the blood, consequently reducing the levels of oxidative stress in the organism [13]. Chauhan et al. (2004) reported that reduced serum ceruloplasmin and transferrin levels are strongly associated with the regression of pre-acquired language skills in autism spectrum disorder children [13].

## 3. OXIDATIVE STRESS IN THE CENTRAL NERVOUS SYSTEM (CNS) OF INDIVIDUALS AFFECTED BY AUTISM

Due to the fact that the brain is most affected in ASD, evidence from neuronal tissue brings more light to the mechanism of oxidative stress in autism spectrum disorder.

GSH, the major cellular antioxidant, has been reported having abnormalities. GSH and GSSG levels in post-mortem brain samples of autistic patients are significantly decreased in the temporal cortex, cerebellum (involved in motor coordination, modulation of cognition and behaviour) and Brodmann area 22 (responsible for speech processing and understanding in dominant hemisphere) as compared with a matched control group (a decrease of 44,6% and 34,2% respectively for temporal lobe and cerebellum) [18, 19].

Several enzymes involved in the redox activity have been studied in order to determine their activity in autistic patients brain. Superoxide dismutase 2 activity has been found decreased in temporal lobe Brodmann area 21 (involved in processing sound and language) tissue samples of 20 autist patients [20]. GPx and Glutathione - S - Transferase have a similar lowered activity in the cerebellum [21].

3-nitrothyrozine (3-NT) is a marker of protein oxidative damage. The pons (involved in autonomic function, eye movements, motor and sensory relay), cerebellum and Wernicke's Area have been found to have decreased levels of 3-NT as compared to healthy subjects [22].

## 4. ROMANIAN PROGRESS IN OXIDATIVE STRESS SCIENCE

In distress, aging and neurodegeneration, etiopathogenic studies demonstrate in the mammalian brain common mechanisms and reciprocal accelerating relationships: hypoanabolism (decrease of RNA and protein synthesis), coupled with hypercatabolism (increase of oxidative stress – lipid peroxidation, protein insolubilizing with waste product accumulation: ceroid – lipofuscin) [23, 24, 25].

Hipopigments (lipofuscin and ceroid) are the main marker of brain vulnerability, distress, aging and connected pathology.

Direct causal interrelations and critical lipopigment concentration, which generate a cascade of negative subcellular events, and indirect, associative impairment correlation, determine characteristic neuropathological aging profiles. These specific and associative negative consequences of lipopigment accumulation have multiple and detrimental impacts on neuron and glia homeostasis, from neuronal function to central nervous system physiology [26].

In opposition, neuronal lipopigments processing, lysis and elimination is one of the main mechanisms for the re-establishment of metabolic, cellular and tissue homeostasis, as well as it is anti-oxidative stress, anti-aging and regenerative therapy – class of the Antagonic – Stress® drugs (Riga D, Riga S – the inventors).

Riga's team (Romania) brain homeostasis therapy is a significant section from the USA – TRANSCEND Research Program Institute: Treatment Research And Neuroscience Evolution of Neurodevelopmental Disorders (www.transcend.research.org, USA)

– Autism Research, Autism Revolution, Reduce Brain Overload, Glial Cells – The Other Brain, A Whole Body Approach to Brain Health [25, 27]. In fact, TRANSCEND are a multimodal and multisystem brain research programs.

Therapeutically activated glia can turn into "brain garbage collectors and transporters". Cells pick up cellular debris, sidle over to blood vessel, and dumps debris into blood vessel. They do this after receiving an intrusive nutritional stimulation program – Riga's (Romania) neurometabolic, neurovascular, anti-oxidative stress and nootropic therapy [27].

## 5. CONCLUSION

Extensive literature evidence reviewed in this paper suggests a major contribution of oxidative stress in autism development, immune, environmental and genetic factor contributing as well. Oxidative stress becomes a pathogenic factor in autism spectrum disorder due to (i) excessive production of endogenous or exogenous oxidant factors, (ii) a decrease of antioxidant capacity or, more frequently, (iii) a combination of both. Several studies on humans or animal models state the abnormalities in the antioxidant enzymes capacity such as superoxide dismutase, glutathione peroxidase or glutathione - S - transferase. Also, increased levels of markers of lipid or protein oxidation, MDA, PUFA resides or 3-NT, outline the oxidative stress state of children with ASD. All these pathological changes, as outlined by the studies reviewed, influence the clinical picture of ASD, determining psychiatric changes and gastrointestinal and sleep disturbances [4].

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