Letters

RESEARCH LETTER

Link Between Increased Prevalence of Autism Spectrum Disorder Syndromes and Oxidative Stress, DNA Methylation, and Imprinting: The Impact of the Environment

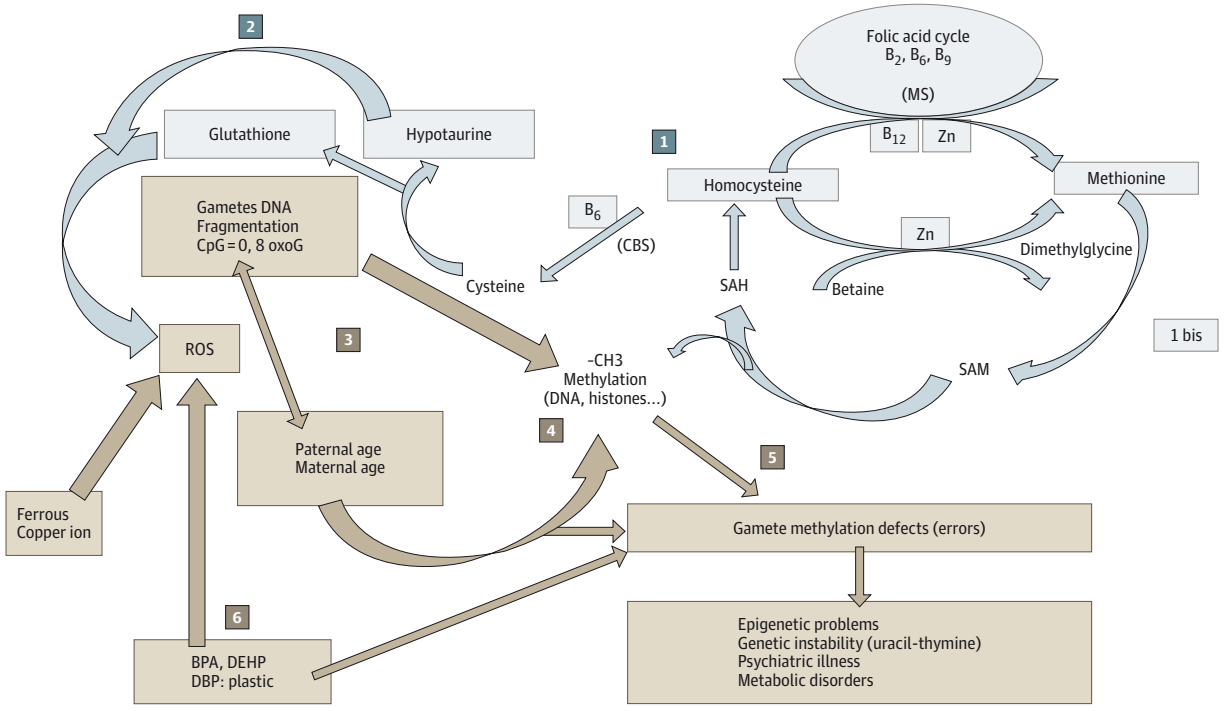
Autism is a complex neurodevelopment disorder, with a male to female prevalence of 4.3:1. The number of children diagnosed with autism or related disorders has increased at an alarming rate: the Centers for Disease Control and Prevention estimates that 1 in 68 children in the United States (or 14.7 per 1000 eight-year-olds) was identified with autism spectrum disorder during 2014. The figure reaches 1 in 45 children in the state of Alabama, and this represents an estimated 30% increase over previous estimates reported in 2012. The prevalence of these disorders has more than doubled since 2000. Here we discuss the biochemical link between the process of DNA methylation in gametes and autism.

Methods | This short commentary is the result of the authors' work on methylation, imprinting, and metabolism in gametes and embryos integrated with the current literature on brain disease and gamete quality.

Results | Disturbances in DNA methylation can originate in the spermatozoon, linked to the age of the male progenitor,[1] and this is expressed as deficiencies in epigenetic mechanisms. Sperm of older men have a higher level of DNA damage, due to a lower resistance to oxidative stress: offspring conceived by older men carrying a high level of sperm DNA fragmentation may escape miscarriage but may instead carry disorders originating from DNA damage that may lead to neuropsychiatric disturbance.

The oocyte expresses folic acid transporters to a high level, whereas cystathionine β-synthase is not expressed, while betaine homocysteine methyltransferase is only weakly expressed.[2] In the absence of an adequate endogenous pool of folic acid in the oocyte, the early embryo's ability to recycle homocysteine

Figure. Interrelations Between Oxidative Stress and Methylation Processes



1, Correct recycling of homocysteine allows generation of cysteine and methionine, which allow correct processes of methylation through the formation of *S*-adenosyl methionine (SAM) (1bis). 2, Correct generation of cysteine allows the synthesis of hypotaurine and glutathione, 2 potent inhibitors of reactive oxygen species (ROS). Hypotaurine is the most important anti-ROS naturally present in vivo in the natural environment of the preimplantation embryo. 3, Generation of ROS induces DNA fragmentation. Advanced age decreases the ability to control ROS-linked decays. 4, High levels of homocysteine perturb DNA methylation processes in sperm, oocytes, and embryos. 5, DNA methylation defects, whether or not linked to imprinting, may result in negative transgenerational health problems. Unrepaired 8 oxoG (oxidized form of guanine) leads to aberrant methylation at CpG sites, which impairs transcription and may affect telomere length (TTAGGG repeats). 6, Plastic derived endocrine disruptors (bisphenol A [BPA], di-[2-ethylhexyl]phthalate [DEHP], and dibutyl phthalate [DBP]) have a negative effect on all of the steps in the pathway.

is handicapped (**Figure**). Intrafollicular homocysteine levels increase in assisted reproductive technologies; therefore, any deficiency in maternal folic acid supplies will affect methylation during very early preimplantation stages of embryo development. Prenatal folic acid supplements have been shown to partially protect against neurodevelopmental disorders in the offspring,[3] as well as have a positive effect on the risk of neural tube defects. A wide range of disorders, including neuropsychiatric disorders, autism, and cognitive impairment, are associated with increased homocysteine levels in biological fluids.[4]

Bisphenol A and other plastic-derived endocrine disruptors have the capacity to inhibit methylation and affect imprinting, inducing epigenetic transgenerational inheritance of metabolic and reproductive disorders, including sperm epimutations.[5] Bisphenol A is a well-known inducer of oxidative stress, as is a high level of circulating glucose. It has been shown that maternal diabetes significantly increases the prevalence of autism in offspring.[6]

Comment | There is therefore a link between methylation and oxidative stress in gametes and the first stages of embryonic development, which potentially effects epigenetic transgenerational transmission. The increase in autism spectrum diseases may also be linked to an increase in environmental endocrine disruptors, which increase oxidative stress and perturb methylation. This effect may manifest in the first 3 days postfertilization up to the blastocyst stage, the period when maintenance of methylation has a significant effect on the imprinting processes, or in the fetus, when imprinting is reset in the germ cells. The sex ratios observed in some disorders may be explained by the higher resistance of female embryos, linked to the *XIAP* gene expression. However, DNA methylation by definition differs between male and female genomes, whether or not it is linked to imprinting; a difference in the sex ratio with respect to autism might therefore be expected. These observations advocate treatment with nutritional supplements that support the 1-carbon cycle for older male and female patients, as well as for female diabetic patients who seek to achieve a pregnancy. The supplementation should include all of the cofactors that contribute to the 1-carbon cycle because, for example, vitamin B_{12} deficiency can induce adverse neurological problems.

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