Role of developmental exposure to environmental agents in altering the disease process is well known. Exposure to chemical agents at critical periods of development may cause some permanent changes in the functioning of various vital systems including the nervous system in the organisms. It is not surprising to see an extensive response due to exposure to chemical agents early in life as the organ systems are more vulnerable to chemical insults during developmental stages. In some cases the response to low level environmental insults may not be obvious until adult or old age. Results from several studies have shown such latency in response to the nervous system leading to neurodegeneration in old age. Studies conducted in murine and primate models provided ample evidence for the association of developmental exposure to low levels of heavy metal lead (Pb) and Alzheimer’s disease-like pathology during senescence. It is not clear about the reasons behind such response; however, the contribution of epigenetic mechanisms could explain the role of early events in life in inducing the late life abnormalities of nervous system. It is possible that environmental agents epigenetically modulate the gene regulation to persist the response silent for a long period of time and to result pathological outcomes significantly later in life. This article will summarize the association of early life exposure to environmental agents and late-life abnormalities with an emphasis on developmental exposure to Pb and neurodegeneration in old age.

Keywords: Developmental exposure, Environmental exposure, Epigenetics, Lead, Neurodegeneration

The role of environmental insults in disease processes is highly defined; however the risk of late life abnormalities due to exposure to environmental agents early in life is not adequately addressed. Barker et al.1 have studied the associations of early life events such as poor nutrition and low birth weights and the incidence of cardiovascular disease in adult age and reported an inverse relationship, thereby showing the first evidence for linking the early life events to adult diseases. Barker and his colleagues’ investigations lead to the hypothesis known as the Fetal Basis of Adult Disease (FeBAD) proposed for the association of many adult diseases with fetal origin1-4. The analysis of clinical and experimental data on diseases of cardiovascular system, Hypothalamic-pituitary-adrenal (HPA) axis and metabolic diseases such as diabetes showed a strong contribution of nutritional imbalances during pregnancy5,6. It was also found that the early life events such as infection, fetal malnutrition and hypoxia may lead to diseases like schizophrenia7,10. It was also proposed that chemical exposure during the early phases of life can impact future disease processes. Studies from various investigators revealed that developmental exposure to some environmental agents (eg., methylmercury, methylazoxymethanol, triethyltin and pesticides) may remain “silent” for a long period of time without any overt manifestations11-15, and suggested that early life events due to environmental insults such as exposure to chemical agents could contribute neurodegenerative manifestations later in life.

Environmental and occupational exposure to myriad chemicals occurs at various stages throughout human life. Many of these chemicals are benign, but some could pose a significant health risk and the exposure to environmental xenobiotic metal lead (Pb) has long been a widespread public concern16,17. The heavy metal Pb has been in use for more than 8000 years. Apart from its application in fuels as an antiknock fuel additive, it has been extensively used in glass, pigments, makeup, water transport pipes, beverages, cooking material, electronics and batteries17,18. Even though organic forms of Pb have been removed from gasoline, inorganic Pb still remains a major environmental hazard facing human populations today. Humans can be exposed to Pb
through paints, glazed earthenware, food containers, beverages (moonshine whiskey), and automobile battery casing\textsuperscript{16,18}. It is well established that developmental exposure to low levels of Pb impacts intellectual functioning and responsible for disturbances in a number of molecular, biochemical and morphological processes\textsuperscript{19-26}. The direct neurotoxic actions of Pb can cause several perturbations including apoptosis, excitotoxicity, disruptions in neurotransmitter storage and release processes, mitochondria and second messengers\textsuperscript{27}. Studies from several laboratories evaluating the role of developmental exposure to Pb and perturbations in cognitive functions revealed that the cognitive impairments due to Pb-exposure may remain for life (reviewed by White \textit{et al.})\textsuperscript{17}. It is not clear whether exposure to Pb during critical periods of development results latent response towards a decline in nervous system functions and/or neurodegeneration during senescence.

**Developmental exposure to Pb and latent responses**

For many decades the Pb-induced toxicity has been mainly considered as a threat for children and the adults with occupational exposure. It is certain that exposure to Pb is an environmental risk facing children, leaving them with residual cognitive and behavioral deficits that persist well into their young adult life; however the ability of developmental exposure to Pb to impact disease processes with long asymptomatic period and explicitly showing clinical symptoms in old age was not addressed until the recent findings reporting the association of developmental Pb-exposure and potential risk of old age neurodegenerative disorders such as Alzheimer’s disease\textsuperscript{16,28-30}.

Alzheimer’s disease (AD) is a progressive neurodegenerative disorder and the most common forms of dementia. The clinical manifestations of this disease usually appear in old age. The hallmark pathological feature of AD is the presence of senile plaques in brain. Amyloid Precursor Protein (APP), is a transmembrane protein and its proteolytic processing generates a 39–42 residue peptides referred to as β-amloid (Aβ) which are the primary constituents of amyloid deposits found in the aging brain and have been implicated in the pathogenesis of AD and Down syndrome\textsuperscript{31-33}. Interestingly, Specificity protein1 (Sp1) transcription factor is associated with the regulation of APP expression and the studies from Zawia and colleagues have identified that Pb-exposure targets Sp1 transcription factor\textsuperscript{22-24,34}. Environmental and other non-genetic factors are causally related to the onset of the more common sporadic forms (~95%) of AD. Twin studies evaluating the inheritance pattern of neurodegenerative diseases including AD and Parkinson Disease (PD) and the sporadic nature of late onset AD suggest a strong role for the environmental influence on the onset of these diseases\textsuperscript{35-37}. Studies on rodents and primates strongly suggest that exposure to Pb during development promotes the pathogenesis of AD\textsuperscript{16,30}. Rodents were lactationally exposed to Pb (0.2% lead acetate in deionized water given to mothers as drinking water) from postnatal day (PND) 1 through 20 (weaning) and brain tissues were collected at various time points from PND 5 through 20 months and monitored the lifetime expression of the AD associated genes. Results revealed that APP mRNA expression was transientsly-induced in neonates, momentarily returned to basal levels but exhibited a delayed over-expression 20 months after exposure to Pb had ceased. This up-regulation in APP mRNA expression was accompanied by an increase in the activity of the transcription factor Sp1. Sp1 is a critical transcription factor associated with the regulators of the APP gene. In consistent with the alterations in APP gene the levels of APP and its amyloidogenic cleavage peptide, Aβ also showed a significant rise during senescence following developmental exposure to Pb (Fig. 1).

![Fig. 1—Early exposure to Pb and latent response. Schematic representation showing effects of development exposure to Pb and its latent effects on expression of genes associated with neurodegeneration](image_url)
(amyloidogenesis and neurodegeneration). Primates are the rare animal models that express amyloid plaques and other pathological features similar to humans and such plaque formation incidences are absent in normal non-transgenic rodents. Experiments using the brain tissue derived from these primates demonstrated that the APP mRNA, APP, and Aβ are elevated in the 23 years old monkeys that were developmentally-exposed to Pb (Birth through 57 week). Immunohistochemical analysis of Aβ deposition shows that early exposure to Pb alters the distribution of intracellular Aβ staining and plaques formation in the frontal association cortex of these animals. The analysis of frontal association cortex of the moneys exposed to Pb during infants showed high abundance of plaques with dense staining for Aβ, while the plaques in control animals were diffused and present in less number (Fig. 2). These studies provided strong evidence that environmental influences (eg., exposure to xenobiotic metal, Pb) during critical periods of brain development could impact the expression and regulation of APP later in life, potentially altering the course of amyloidogenesis, thereby acting as a risk factors for the onset of AD-like pathology in rodents and primates in old age.

Developmental Pb-exposure and oxidative DNA damage

It is well established that oxidative damage to DNA is associated with neurodegenerative diseases. As mentioned above neonatal exposure to Pb resulted over expression of APP and Aβ peptides in old age. Since Aβ peptides are cytotoxic, induction the generation of reactive oxygen species and promote neurodegeneration in the aging brain, it is important to corroborate whether early exposure to Pb impacts the onset of oxidative damage late in life. Bolin et al. studied the levels of 8-hydroxy-2′-deoxyguanosine (oxo8dG) as a measure for the accumulation of oxidative DNA damage and the activity of DNA repair enzyme 8-oxoguanine DNA glycosylase (Ogg1) in the brain tissue (cerebral cortex) of 20 months old rats following neonatal (PND1-20) and old age (18-20 months) exposure to Pb. Oxo8dG was transiently modulated at PND 5 but returned to basal levels on subsequent time points and abruptly showed an elevation 20 months after exposure to Pb had ceased. On the other hand the Ogg1 activity was unaltered. The effect of Pb on oxo8dG levels did not occur when animals were exposed to Pb in old age (18-20 month). Bolin et al. also evaluated the changes in super oxide dismutase (SOD) enzymes and reduced-form glutathion (GSH) which are associated with antioxidant mechanism. The production of reactive oxidative species (ROS) due to environmental insults instigates DNA damage causing strand breaks and base oxidation in DNA. SOD1 and SOD2 represent a family of enzymatic antioxidant defense involved in converting superoxide into peroxide and act as defense mechanism against oxidative stress. The increases in DNA damage due to neonatal exposure to Pb failed to produce changes in copper/zinc-superoxide dismutase (SOD1), manganese-SOD (SOD2), and reduced-form GSH. These data strongly suggest developmental disturbances such as chemical injury to nervous system potentially impacts oxidative damage and 

![Control](image1.png) ![Pb-exposed](image2.png)

Fig. 2—Developmental exposure to Pb increases plaque formation in primates. Images showing the AD-like pathology in the frontal association cortex of 23-year-old cynomolgus monkeys after developmental exposure to Pb.
neurodegeneration in the aging brain. The association of oxidative DNA damage and alterations in methylation patterns due to developmental exposure to Pb is described below.

### Epigenetics

Epigenetics refers to the mechanism that causes changes in phenotype or gene expression without change in DNA sequence. The molecular basis of epigenetics is a complex phenomenon and involves modifications of the activation of genes due to chromatin remodeling through posttranslational histone modifications and/or alterations in DNA methylation. It is proposed that the permanent changes or long-term alterations in gene expression may be due to changes in DNA methylation patterns induced by developmental exposure to chemical agents. Environmental agents could impact DNA methylation through disrupting the enzymes involved in methylation reactions. Studies from other laboratories showed that cadmium (Cd) treatment can alter DNA methylation pattern of certain genes in consistent with the changes in the activity of methylation catalytic enzyme DNA-methyltransferase. To determine whether developmental exposure to Pb interferes with DNA methylation patterns, the activity of DNA-methyltransferase was monitored in the tissues of 20 month old rodents that were exposed to Pb from birth through weaning (PND 1-20) and 23-year-old primates that were exposed to Pb as infants and found that this enzyme is highly diminished in Pb-exposed animals. It is clear from the studies on rodents and primates that the developmental exposure Pb enhanced the expression of genes associated with AD and increased the burden of oxidative DNA damage in the aged brain. It is also clear that epigenetic mechanisms that control gene expression and promote the accumulation of oxidative DNA damage are mediated through alterations in the methylation or oxidation of CpG dinucleotides. It is possible that the environmental influences such as chemical insult during brain development can inhibit DNA-methyltransferases, thereby hypomethylating promoters of genes associated with AD (eg., APP, BACE1) and such early life events may imprint and sustain to trigger the process leading an increase in gene expression. The increased expression of APP gene ultimately leads to an increase in the production and accumulation of β-amyloid peptides (amyloidogenesis) in brain. The amyloidogenesis will promote the formation of reactive oxygen species, which damage DNA and accelerate neurodegenerative events. The changes in hypo or hyper methylation of genes can impact on their expression and imprint susceptibility to oxidative DNA damage in the aged brain. It is plausible that developmental exposure to Pb could exacerbate the demethylation process of the APP promoter in old age and elevates its expression. The hypothesis to explain how environmental insults such as exposure to Pb can disturb the expression of disease (neurodegeneration) associated genes through impacting the methylation process is schematically illustrated in Fig. 3.

### Conclusion

The studies showing the association of developmental exposure to Pb and AD-like pathology in old age argues for the potential risk of late life abnormalities due to injury/insult to vital systems such as nervous system early in life. It is also evident that Pb acts through a variety of mechanisms to affect developmental processes, neurotransmission, behavioral and cognitive functions and some of these alterations may not obvious until old age suggesting the latent response and potential risk for the development of neurodegenration in old age due to alterations in the course of amyloidogenesis.
References


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