Toxic Metals: Health Effects, and Therapeutic Measures

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ABSTRACT

Metal intoxication, thus is an important concern in the world today owing to high industrial and chemical exposure to humans. These metal toxicities have been extensively studies with underlying mechanisms revealed up to molecular levels. However, much emphasis and focus on managing these metal-induced toxic manifestations is the need of the hour. Chelation therapy has long been surrounded with debates and challenges for unanimous acceptance despite being the only option. Although conventionally known chelating agents show therapeutic efficacy, yet more specific and safer profile drugs are needed. Newer therapeutic strategies like combination therapy (administration of two chelating agents) or co-administration of chelating agent with antioxidants and/or essential metals needs to be clinically refined and implemented.

Keywords: Toxic Metals; Toxicity and health effects; Mechanism of action; Preventive and therapeutic measures

INTRODUCTION

Metals form inseparable segment of our lives and living biological system. These may be essential for life, facilitate biological pathways or interfere to inhibit the same. Besides their great applicability metals comprehensively influence all life forms causing severe effects commonly designated as “Metals induced toxicity”. Metals as environmental and occupational toxicants are gaining concern for their toxic manifestations in humans. Increasing industrial use of metals has led to an environment in which chronic intoxication is common. Heavy metals in particular, form one of the most hazardous environmental toxicants posing deleterious health risk in humans due to continue exposure. Researchers dedicated to investigate metal toxicity have achieved considerable success in analysing risk assessments in humans, identifying the diagnostic markers and revealing the underlying pathways. In this chapter we provide an insight to toxicity of some of the common and highly toxic metal such as Aluminium, Arsenic, Lead, Mercury, Cadmium, Iron, Chromium, Nickel, Manganese, Platinum and Thallium, along with a brief review on metal chelation.

TOXIC METALS

ARSENIC

The word arsenic is derived from a Greek word arsenikon, which itself is derived from a Persian word Zarnikh, meaning yellow orpiment. Although it is a metalloid with characteristics of both metals and nonmetals, arsenic is commonly characterized as a heavy metal. Arsenic is highly toxic, which poses severe effects and its use as a deadly poison has been known and reported for many years. Due to its use by the ruling class to murder one another\(^1\) and its potent effects, arsenic has been called as Poison of Kings and the King of Poisons.\(^2,6\) During world war I and Vietnam war it was used as chemical warfare agent named as lewisite (ClCH=CHAsCl\(^2\)), and Agent blue acting as a vesicant (blister agent) and lung irritant. Besides being toxic, arsenic holds an important position in traditional medicinal therapies in China, India,\(^3,8\) along with Greece and Rome.\(^9\) Recently it has been used as a treatment for late-stage African trypanosomiasis (mefloquine) caused by Trypanosoma gambiense or T. rhodesiense and for acute promyelocytic leukemia as arsenic trioxide, marketed as Trisenox\(^8,11\).

Environmental contamination of arsenic particularly in drinking water sources mainly because of anthropogenic activities, is a major cause for concern in many parts of the world. Reports of large-scale arsenic contamination in the
Gangetic Delta region in Bangladesh and India have drawn significant attention. In this part of the world alone, more than 38 million people are at risk of developing arsenic related health hazards. World Health Organization recommends maximum permissible value for arsenic in drinking water to be 10 ppb. However, many countries like Argentina (200 ppb), Mexico (400 ppb), and the Indo-Bangladesh region (800 ppb) have extremely higher arsenic concentration in drinking water.12

Naturally arsenic occurs in rocks, soil, metal ores (such as copper and lead), and in the form of minerals. The two most common minerals of arsenic are arsenopyrite and loellingite, from which arsenic is isolated through smelting producing elemental arsenic. Commercially arsenic is produced as elemental arsenic and arsenic trioxide (As$_2$O$_3$). Compounds derived from arsenic have a number of commercial applications such as catalysts, bactericides, pesticides (eg. lead arsenate, calcium arsenate, and sodium arsenite), herbicides (eg. mono sodium arsenate and dimethyl arsenic acid), cotton desiccants (eg. arsenic acid), wood preservatives (eg. zinc and chromium arsenate), fungicides, animal feed additives, corrosion inhibitors, veterinary medicines, and tanning agents. Human exposure to arsenic can occur from inhalation of particulate arsenic, or absorption through the skin. Commonly arsenic exposure occurs by ingestion of water and food particularly seafood, rice, mushrooms, and poultry, which are reported to have the highest concentrations of arsenic.13 Air, water, and arsenic containing mineral ores represent some common environmental source of arsenic exposure. However, the main source of human environmental exposure is through consumption of water containing elevated levels of arsenic, primarily from natural contamination.14

Arsenic induces toxicity following its metabolism which is a two step procedure. Oxidation from trivalent to pentavalent or reduction from pentavalent to trivalent by a common enzyme arsenate reductase.15,16 In precision to this, arsenite is further metabolized from inorganic to organic form through sequential methylation reactions. During second step monomethylarsenic acid (MMA) and dimethylarsinic acid (DMA) are formed in vivo17 using S-adenosyl methionine (SAM) as the methyl donor and GSH as an essential co-factor. Both MMA and DMA are more toxic than the parent compound and cause serious effects including cancer.6,18,19 MMA and DMA can be further reduced into the more toxic trivalent form.6,17,18

Arsenic, particularly trivalent forms, binds to sulphydryl groups, disrupts essential metals, and leads to impaired gluconeogenesis and oxidative phosphorylation. For example, in glycolysis one of the intermediate product is 1,3-diphosphoglycerate which produced from glyceraldehyde 3-phosphate. The product undergoes subsequent reactions to produce ATP, which is the energy currency of the cell. When arsenite AsO$_3^{3-}$ is present, it binds to glyceraldehyde 3-phosphate to yield a product that spontaneously undergoes non-enzymatic hydrolysis, thereby preventing ATP formation (Figure 1).

Figure 1. Arsenite interference in production of ATP by phosphorylation.

In another mechanism arsenic inhibits the enzyme pyruvate dehydrogenase, by binding to its sulphydryl groups. This inhibits conversion of pyruvate to acetyl coenzyme A (CoA) thereby inhibiting ATP biosynthesis as acetyl coenzyme A (CoA) is the precursor of Kreb’s cycle which is the driving source to generate ATP. Beside this arsenic also inhibits the uptake of glucose into cells, gluconeogenesis, fatty acid oxidation, and further production of acetyl-CoA.

The toxicity of pentavalent inorganic arsenic (iAs) occurs by two ways, one via its reduction to trivalent arsenic. Secondly due to its resemblance with inorganic phosphate, it gets substituted in place of phosphate in glycolytic and cellular respiration pathways. This results in preferential formation of ADP-arsenate that has less stable bonds in comparison to high energy compounds such as ATP. Thus, situation termed as ‘arsenolysis’ causes a rapid hydrolysis of these bonds, resulting in premature uncoupling oxidative phosphorylation.

Arsenic-induced oxidative stress is considered an important mechanism for its toxicity.12 Generation of various types of ROS during arsenic metabolism in cells has been confirmed.20 In addition to ROS, reactive nitrogen species (RNS) are also believed to be directly associated in oxidative damage to lipids, proteins and DNA in cells exposed to arsenic.21 Superoxide anion radicals like (O$^2_-$), singlet oxygen ($^1$O$_2$), the peroxyl radical (ROO$^-$), nitric oxide (NO), hydrogen peroxide (H$_2$O$_2$), hydroxyl radicals,22 dimethylarsinic peroxyl radicals ((CH$_3$)$_2$AsOO) and also the dimethylarsinic radical [(CH$_3$)$_2$As] are generated through reactions involved arsenic.23 However, the exact mechanism for the generation of these reactive species is not clear, but it is proposed that there formation involves intermediary arsine species. GSH, an effective cellular antioxidant is reported to reduce or elevate during arsenic exposure.24,25 These trends are dose and duration of arsenic exposure dependent. GSH possibly acts as an electron donor for the reduction of pentavalent to trivalent arsencicals and that arsenite has high affinity to GSH, rendering it unavailable for subsequent reactions. These effects in combination disturb cellular homeostasis causing different cell malfunctions including cancer. Many recent studies have provided experimental evidences which show that arsenic exposure has been linked with various types of cancer.26 cardiovascular
disease, diabetes, neurological disorders and dermal effects.

Symptoms of acute arsenic poisoning are many and may be severe - fatal at high doses. Severe cases of arsenic poisoning have exhibited symptoms of fever, aversion to food, abnormal liver enlargement (hepatomegaly), cardiac arrhythmia, development of dark patches on skin and other tissue (melanosis), peripheral neuropathy, including sensory loss in the peripheral nervous system, gastrointestinal disorders, cardiovascular effects, and adverse effects on red blood cell formation, which can result in anemia.

Chronic effects of arsenic poisoning include neurotoxic effects to the central and peripheral nervous systems. Liver injury is a common symptom of chronic arsenic poisoning. Studies of victims of chronic arsenic poisoning from contaminated drinking water in Taiwan and Chile have exhibited blueness of the skin in extremities, a condition called acrocyanosis, the result of peripheral vascular disease. In extreme cases, this may progress to gangrene in the lower extremities, a condition called Blackfoot disease.

**CADMIUM**

Cadmium is the 48\textsuperscript{th} element and a member of group 12 in the periodic table of element, with +2 as the most common oxidation state. It is a wide spread metal contaminating many areas, either naturally or by anthropologic activities, well recognized as a health hazard. The famous itai-itai (“ouch-ouch”) disease of Japan characterized by multiple fracture and distortion of the long bones, and severe joint and spine pain was attributed to consuming of cadmium-polluted rice. Cadmium has been listed amongst the 126 priority pollutants by US Environmental Protection Agency (EPA), and has been classified in number one category human-carcinogen by International Agency for Research on Cancer of USA.

Cadmium is virtually present in all natural sources of food. High concentration of cadmium is generally observed in sea foods such as molluscs, crustaceans, cephalopods and crabs; oil seeds; cocoa beans; certain wild mushrooms; animal products like liver and kidney of exposed animal; plant products like cereals, green leafy vegetables, potato, carrot, and tobacco. It has been estimated that more than 80% of dietary cadmium comes from cereals, vegetables and potato. Industrial sources of cadmium loading are mainly electroplating, smelting and refining, welding, pigment production, and battery manufacturing industries. Although the main cause of cadmium loading occurs via ingestion of contaminated food or water, inhalational exposure also occurs through cigarette smoke, indoor cadmium contaminated dust or occupational exposures. Once absorbed, cadmium binds to albumin and is transported to liver, where it promotes the synthesis of metallothionein (MT), a small cysteine-rich heavy metal-binding protein. The MT–cadmium complex is then released from liver to plasma and eliminated in urine. MT-bound cadmium can be reabsorbed from the glomerular filtrate by the renal tubule cells, where it is cleaved by lysosomal action, thus releasing Cd\textsuperscript{2+} ions that are re-excreted into the tubular fluid. This free cadmium is then released in the cytoplasm where it binds rapidly to intracellular protein including GSH and MT.

Cadmium toxicity is mainly associated with generation of tumors. In cadmium-related carcinogenesis, various regulatory genes are activated including ‘Immediate early response genes’ (IEGs). IEGs, involved in cell proliferation and differentiation are often over-expressed in tumors. IEG over-expression constitutes mitogenic growth signals stimulating cell proliferation and induction of carcinogenesis. Another target for cadmium-induced carcinogenicity is induction of several stress response gene expression such as those encoding synthesis of metallothionein (MT), heat-shock proteins (HSPs), reduced glutathione (GSH), genes involved in oxidative stress response, etc. at cellular level, cadmium is found to influence several transcription factors and translational genes. It is a powerful inducer of c-fos and c-jun which has been identified as a major mechanism for cadmium-induced cell transformation and tumourigenesis. Cadmium directly generate free radicals itself and indirectly via involving superoxide radical, hydroxyl radical and nitric oxide. Generation of non-radical hydrogen peroxide, which itself may serves as a significant source of radicals via Fenton’s chemistry has also been reported. Other cellular effects of this toxic metal are induced by disruption of physiological signal transduction systems, including those mediated by Ca\textsuperscript{2+}, cAMP, NO, MAP-kinase, PKB/Akt and nuclear factor-kappa-B.

Cadmium-induced malfunctioning at molecular and cellular level ultimately leads to various disorders including pulmonary edema, hemorrhage, fulminate hepatitis, testicular injury. At high concentrations its toxic symptoms include renal damage characterized by early increase in excretion of low molecular weight proteins (β2 and α1 microglobulins) due to glomerular damage and dysfunctioning of tubular reabsorption, along with glycosuria and aminoaciduria.

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**Figure 2.** Toxic effects of cadmium at molecular level.

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Chromium (Cr) is the 24th element of the periodic table and is 21st most abundant element in the Earth’s crust at about 100 ppm. It is ubiquitously present in the environment due to erosion of rocks, volcanic eruptions, soil, sea water and rivers. Chromium has been in demand since the discovery of Crocoite, the first ore of lead-chromate till today owing to the versatile application and the component of stainless steel. Chromium also finds its application in other industries including chromate production, chromate pigment production for metal, glass and synthetic rubies, chrome plating, preservation of wood, tanning of leather, refractory material, super alloys for jet engines and gas turbines etc. Chromium exists in two important stable states trivalent [Cr (III)] and hexavalent [Cr (VI)]. Cr (III) is an essential micronutrient for biological activity of insulin, glucose and lipid metabolism and its deficiency has been associated with impaired glucose tolerance, hyperglycemia, glucocuria, diabetes, cardiovascular disease etc. It is found in most fresh foods, vegetables, cereals, spices, bread and drinking water. Chromium naturally exists in trivalent [Cr (III)] state which is non-carcinogenic, while the hazardous hexavalent [Cr (VI)] form is predominantly produced by anthropogenic activities. Human exposure to chromium occurs via inhalation due to occupational (industries) or non-occupational (automobile emissions and smoking cigarettes) sources.

Chromium plays the most controversial role in terms of its biological activities. Cr (III) is nontoxic by virtue of its inability to pass through cell membrane, while Cr (VI) is transported inside the cells through anion channels as chromate. Once Cr (IV) ions enter the body they are spontaneously and passively absorbed by cells. It then undergoes rapid metabolic reduction in presence of cellular reductants such as ascorbic acid, reduced glutathione (GSH) and cysteine to generate stable Cr (III) and unstable Cr (IV) and Cr (V) intermediates. During the reduction, Cr (VI) generates reactive oxygen species (ROS) by Fenton and Haber-Weiss type of reactions which ultimately results in oxidative stress causing DNA lesions including Cr-DNA adducts, DNA-protein crosslinks, DNA-DNA crosslinks, activation of nuclear transcription factors, up-regulation of antioxidants, and activation of enzymes responsible for Cr (VI) reduction. Reduction of Cr (IV) also induces cell cycle arrest at G1 phase, S-phase, and G2 phase. Cr (IV) induces apoptosis by activating both intrinsic mitochondrial pathway and extrinsic death factor pathway. In addition to cancer, chromium-induced toxic manifestations include dermatitis, hand ulcers, perforation of the nasal septum, renal and hepatic.

Mercury is considered one of the most toxic pollutants in the environment belonging to transition metals at 80th position in the periodic table. Mercury although is a liquid, it records for the highest density and is listed as a heavy metal exhibiting...
toxicity at varied levels. Mercury exists as cations in two oxidation states of +1 (mercurous) or +2 (mercuric). Methylmercury is the most encountered form of organic mercury formed from methylation of inorganic mercury through the action of anaerobic organism that live in soil and water. All forms of mercury including elemental (Hg), inorganic (mercurous, Hg(I) and mercuric, Hg(II)) and organic mercuric compounds are ubiquitous for possible human exposure and show toxic effects. Emanation of mercury in the environment occurs naturally from volcanic emission, oceanic sediments, degassing from geological materials and by forest fires, whereas anthropogenic sources includes industrial uses, burning of fossil fuels, incineration, mining and from degradation of mercury containing compounds. Humans subjected to mercury poisoning due to occupational exposure or through contaminated food, are at risk of severe disorders like neurological alterations affecting cognitive and motor dysfunctions, tremor, mental disorders, ataxia, disturbance of taste and smell, spasticity and blindness. Methylation of mercury by microorganisms such as Methanobacterium greatly increases the transport of mercury in aquatic animals and thus, ultimately leads to its deposition in humans.

The general mechanism of toxicity involves the covalent binding of mercury with sulfhydryl groups inactivating various enzymes affecting cellular functioning and metabolism. Besides higher affinity for sulfhydryl groups, it also binds to primary and secondary amine, amide, carboxyl and phosphoryl groups. Extent of mercury induced toxicity is determined by its chemical form. Elemental mercury vapours are highly lipid soluble, predominantly absorbed through lung, crosses the alveolar membrane and easily reaches systemic circulation and body tissue. Following systemic distribution mercury rapidly undergoes oxidation to mercuric ions and thereby binds to ligands in the red blood cells (RBCs). Thus reaching brain it leads to progressive CNS dysfunction and pathological alteration of cellular membrane. Elemental mercury is eliminated from the body as mercuric ions by the urine and faeces following a biphasic elimination rate, initially rapid, then slow. Elemental mercury vapours are easily transported across the placenta. Organic mercury is lipid soluble and is rapidly absorbed through inhalation, ingestion or dermal exposure. More than 90% of methyl mercury binds to erythrocytes and slowly distributes and accumulates in the liver, kidney, brain, hair and epidermis. In humans, the presence of body’s methylmercury burden is nearly 10% in the brain with approximately 70 days half-life. Methylmercury readily crosses blood-brain barrier and reaches all areas of the brain through mechanisms that are not fully characterized. In humans, its main route of excretion is through the faeces, with less than 10% in the urine and is also excreted in bile. It can cross the placenta, accumulates in the foetus and excreted in toxic amount in breast milk. Inorganic mercury poisoning occurs as a result of intentional or accidental ingestion. Mercury salts are usually absorbed through the gastrointestinal tract in dose dependent manner but exhibit low bioavailability (5-10%) as compare to organic compounds. After absorption, the salt dissociates into the ionic form, distributed between RBCs and plasma, and then reaches the tissues affecting the gastrointestinal tract and kidneys. Distribution of mercury within the body and tissues varies greatly and accumulates predominantly in the renal cortex. Autoradiographic study revealed that the mercury uptake by brain is slow but is retained for a longer duration than kidney. Rate of excretion of mercury is dose dependent and is readily eliminated by urinary and faecal routes.

Metallothionein play a crucial role in scavenging and reducing the toxic effects of mercury due to the presence of sulphydryl groups. MT is induced not only in cadmium and mercury but also with other metals like Zn and Cu. Exposure to mercury (organic or inorganic) induces oxidative stress in experimental animals mainly because of depletion cellular thios, especially GSH, lipid peroxidation and increased formation of H2O2 in the kidneys of rats. These oxidative stress responses are found to be mediated through mitochondria of renal cells in vivo and in vitro models.

Mercury also induces developmental toxicities with predominant neurotoxicological effects. Mice offsprings were found more susceptible to mercury-induced neurotoxicity mimicking those in humans. The signalling of ROS/Na+-ATPase/NOx is found to play a crucial role in the underlying mechanism for mercury-induced toxic effects in offspring.

Further, recent findings on methylmercury-induced neurotoxicity and cell death pathways have been described in neural and endocrine system by disrupting calcium homeostasis, induction of oxidative stress via generation of ROS by parallel reduction of antioxidative enzymes and interaction with sulphydryl groups.

**Thallium:**

Thallium (Ti) is a naturally occurring, highly toxic heavy metal belonging to group III A of the periodic table. Although it is toxic in nature, very few studies have been reported due to lack of appropriate, reliable and sensitive method for its detection. US EPA included thallium in the list of priority toxic pollutants’ owing to the fact that it is responsible for a number of occupational and accidental poisonings. It is introduced into the environment mainly as waste from the production of zinc, cadmium and lead and by combustion of coal. Thallium is extensively distributed in the earth’s crust in the form of minerals and salts and is readily released and transported with alkaline metals during weathering process. Anthropogenic sources of thallium pollution are gaseous emissions from cement industries, coal based power plants, and metal sewers. Leaching of thallium from ore processing operations is the major cause of thallium contamination in natural water sources. Other major sources of thallium include industries of copper smelting, petroleum refining, nonferrous metals, blast furnaces, and steelworks. Thallium exposure to humans can occur through air, water and food. Exposure occurs through ingestion of food contaminated with thallium while air and water concentrations are low in thallium levels. Inhalational and dermal exposure also results in thallium absorption.

Clinical manifestations of thallium poisoning can be characterised as acute, sub-chronic, and chronic, depending on
the dose of thallium, severity, route and duration of exposure. The most significant symptom of thallium poisoning is the loss of hair or alopecia. Acute thallium poisoning usually results in gastrointestinal symptoms, while neurological findings such as sensory and motor changes predominate in chronic exposure. Other symptoms include polyneuritis, encephalopathy, tachycardia, degenerative changes of the heart, liver, and kidney, sub-archanoid haemorrhage, and bone marrow depression. Exposure to thallium for about one month leads to formation of transverse white lines called as “Mee’s lines” in the nail plate due to erosion of the proximal parts of nails following thallium poisoning.

Thallium is a highly toxic metal and exhibits adverse effects on various organ systems. Although precise mechanisms involved in thallium toxicity is unclear yet thallium is shown to induce toxicity to living cells by (1) altering the biological processes involving potassium ions; (2) inducing oxidative stress; (3) per-oxidation of lipids there by disrupting membrane permeability and affecting membrane bound enzyme activities; (4) disturbing mitochondrial function and (5) exerting neurotoxicity. Thallium due to its same ionic radius as that of potassium (K⁺ and Tl⁺ ions) replaces K⁺ ions and follows potassium distribution pathways. One such example is replacement of K⁺ in Na⁺K⁺-ATPase by Tl⁺ which has ten times greater affinity than that of K⁺. Thallium also binds to the thiol groups thus inhibiting various relevant enzymes and depleting glutathione leading to oxidative stress condition. It may also bind to the membrane phospholipids and thereby altering the fluidity of the membrane, lipid packing, permeability of the membrane and activity of the membrane bound enzymes and receptors. Thallium in millimolar concentrations affects the mitochondrial function by uncoupling the respiratory chain and by opening the transition pores. It was also reported to cause swelling of mitochondria and increased oxygen consumption and lactic acid production in vitro.

Thallium toxicity results in symptoms like gastrointestinal disturbances, anorexia, abdominal and retrosternal pain, vomiting, constipation, polyneuritis, darkening of hair roots, insomnia, alopecia and alteration in blood pressure. In severe cases, tachycardia, hypotension, hyperflexia and peripheral cyanosis can also be noticed. Thallium even at low doses may result in gastrointestinal haemorrhage, gastroenteritis, metallic taste, salivation, nausea, and vomiting, followed by neurological disorders, hallucination, lethargy, delirium, convulsions, tingling pain in extremities, and muscular weaknesses including coma. Thallium-induced death arises due to respiratory and cardiac failure.

Effect of thallium on sweat and sebaceous glands causes palmar erythema, acne, anhidrosis and dry scaly skin. Neurological symptoms usually appear between 2–5 days after acute exposure cases, which are characterized by a painful, rapidly progressive peripheral neuropathy that dominates clinically in the second or third week. Sensory disturbances include pain and paresthesias of the lower limbs, numbness in the fingers and toes, with the loss of pin-prick and touch sensation. Motor neuropathy is evident by weakness which is always distal in distribution. The lower body extremities are primarily affected. Upper extremity involvement occurs uncommonly and cranial nerves participation is rare. Insomnia, headache, emotional liability, anxiety, tremor, ataxia, choreoathetosis and signs of cranial nerve involvement may be developed. Psychosis with paranoia, depression, aggressiveness and hallucinations are also common. In chronic poisoning, ataxia and paresthesia may be the outstanding symptoms. In time, the paresthesia may progress to evident peripheral neuropathy with weakness and atrophy of the associated musculature. In very serious or even fatal cases, true ‘pseudobulbar paralysis’ due to peripheral neuritis of cranial nerves is observed, with paralysis of the ocular muscles, facial paralysis, ambyopia and paralysis of the recurrent nerve. Shortly before death, paralysis of the vagal nerve may supervene, possibly being the direct cause of death. With some cases, in early stages of poisoning, the optic disk reveals the typical picture of neuritis with ill-defined and red papillae, followed by the development of pale or white papillae as a result of atrophy of the optic nerve. In occasional instances, the initial stimulation of the ganglionic cells of the brain may give rise to severe Jacksonian epileptic seizures; also severe epileptic form convulsions can be observed.

Cardiac signs, such as a sinus tachycardia, irregular pulse, hypertension and angina-like pain, have been reported following ingestion of thallium sulfate. Some authors suggested that such signs are due to vagus involvement, while others have recorder electrocardiographic evidence of myocardial damage. The most dreaded effect of bilateral paralysis of the vagus nerve is malfunction of the cardiac muscle evidenced by tachycardia and circulatory disturbances, possibly resulting in left-sided (cardiac asthma) or right sided decompenation. Under these circumstances, it is quite common for paralyzed patients, often short of breath already, to develop severe dyspnoea and cyanosis, followed by death. Renal excretion of thallium sulfate is slow and may be detected in months after ingestion. Toxic injures to the kidney have been indicated by albuminuria and haematuria, however, the renal function is not grossly impaired.

Reproductive system is highly susceptible to thallium. Chronic exposure to thallium resulted in decreased libido and impotence in humans. Thallium is accumulated in high levels in testes of human and animals. Morphological and biochemical changes in the testes and decreased epididymal sperm motility has been observed in rats exposed to 10 ppm of thallium in drinking water of 2 months. Since thallium cross blood placental barrier, human foetus may suffer from transplacental exposure, as evidenced by skin and nail dystrophy, alopecia and low body weights in newborns of intoxicated mothers; whereas in animals, thallium also can be detected in fetal tissues after placental transfer of thallium. Thallium is reported to be teratogenic in chick embryos, resulting in achondroplasia, leg bone curvature, parrot beak deformity, microcephaly and decreased fetal size. Nonossification of the phalanges and vertebral bodies reported in mice of thallium exposed mother. When intoxication occurs after first 3 months, only a few newborns showed the symptoms of thallotoxicosis. Exposure to thallium at high levels of the mother at the end of pregnancy leads to the death of the fetuses.
LEAD

Lead is a bluish or silver-grey metal, and is a well-reputed toxin that has been a part of human life from thousands of years. From its application in “pencils to gasoline to kitchen appliances”, the use of lead has been inevitable, making human exposure to lead unavoidable. Some common symptoms appearing at various stages and doses of lead are tabulated in the following table.  

Measuring of blood lead concentration is the most effective and accepted diagnosis for lead exposure. The accepted toxic threshold for lead in infants, children and women of child bearing age is ≤ 10 µg/dL, approved by American Pediatric Association. However, for adults there is no such threshold, as concentration of lead from 10 µg/dL and above in blood exhibits toxicity. Moreover quantitative estimation protocols for detection of lead in the range of 10-30 µg/dL is very low, thus now a day’s measuring blood lead is not recommended for its screening.

Being a divalent cation, lead has tendency to attain entropically stable conformation, to accomplish its stable conformation lead binds covalently with sulfhydryl group. This causes disturbances in enzymatic activities and other protein malfunctioning.

Table 1. Symptoms of lead toxicity

<table>
<thead>
<tr>
<th>Earliest Symptoms</th>
<th>Symptoms of chronic exposure</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Diffuse muscle weakness.</td>
<td>• Abdominal pain/cramping.</td>
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<tr>
<td>• General fatigue/lethargy.</td>
<td>• Nausea/vomiting.</td>
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<tr>
<td>• Myalgia.</td>
<td>• Short-term memory loss.</td>
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<tr>
<td>• Joint pain/arthritis.</td>
<td>• Depression.</td>
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<tr>
<td>• Loss of appetite.</td>
<td>• Inco-ordination.</td>
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<tr>
<td>• Unusual taste in mouth/Change in taste of food.</td>
<td>• Numbness and tingling in extremities.</td>
</tr>
<tr>
<td>• Headache.</td>
<td>• Constipation.</td>
</tr>
<tr>
<td>• Insomnia.</td>
<td>• Inability to concentrate.</td>
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<tr>
<td>• Irritability.</td>
<td>• Impotence.</td>
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<tr>
<td>• Diminished libido.</td>
<td>•...</td>
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<tr>
<td>• Weight loss of 10 lbs or more without known cause tremulousness.</td>
<td></td>
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<tr>
<td>• Personality changes</td>
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</tr>
</tbody>
</table>

Severe toxicity (when the blood lead is elevated over 30 µg/dL)

• Frank paralysis
• Somnolence/severe lethargy
• Abdominal colic

The most common of such interference involves that in haemesynthetic pathway, where it specifically reduces/inhibits the level of enzyme delta aminolevulinic acid dehydratase (delta-ALAD). Thus ALAD is identified as a reliable biomarker for lead exposure since 50% of ALAD inhibition has been account to occur at estimated blood lead levels of 20 µg/dL. However, lowering of ALAD may also be due to other factors such as alcoholism, liver cirrhosis, porphyria, etc. Hence, it may also be misleading in certain cases for detection of lead toxicity. Other diagnostic methods include radiological examination of bones for the presence of “Lead lines” along metaphyses of long bones and along the margin of flat bones; nerve conduction velocity testing, which is indicative of cognitive function impairment caused due to lead toxicity when blood levels are found to be above 80 µg/dL.

Lead toxicity results in a variety of physiological, biochemical, and behavioral malfunctioning which mainly are associated with central and peripheral nervous system, haemopoetic system, cardiovascular system, liver, kidney, and reproductive system. In heme biosynthesis pathway lead inhibits ALAD and ferrochelatase, the two regulatory enzymes resulting in accumulation of ALA. High concentration of ALA induces ROS generation. It is believed that ALA tautomerises to ALA-enol form which donates electron to molecular oxygen. On the other hand as a course property of exchange of gases in respiration oxy-haemoglobin also transfer electron to oxygen.

Figure 4. Sources and metabolism of lead

This cascade of electron transfer produces $H_2O_2$ and $O_2•- which upon interacting with each other produce HO• radicals, one of the strongest known ROS. The hydroxyl radicals in turn may react with cystiene-containing proteins forming thyl radicals. These radicals oxidize GSH and produce superoxide ions. Fuchs et al demonstrated that 4,5-dioxyvaleric acid, the final oxidation product of ALA alkylates the guanine moieties of DNA causing lead induced genotoxic effects. Lead also hinders the functioning of various antioxidant enzymes like, catalase, superoxide dismutase (SOD), glutathione peroxidase (GPx), glucose-6-Phosphate dehydrogenase, and GSH like antioxidant. Glucose-6-Phosphate dehydrogenase is thiol containing regulating enzyme of pentose phosphate pathway, the cell’s generator of reducing equivalence producing NADPH used to maintain equilibrium between GSSG and GSH. As RBC’s are devoid of mitochondria, lead induced inhibition of pentose phosphate pathway makes them sensitive to oxidative damage and premature degradation of RBC, causing anemia. Studies have shown that neurotoxic behavior of lead is due to...
either inhibition of K^+ stimulated release of γ-aminobutyric acid (GABA) or GABA binding to synaptic membrane. Lead induces ROS mediated increase in intracellular level of Ca^{2+}, causing depression of mitochondrial potential, leading to cytochrome c mediated apoptosis. Other potentially toxic effects of lead include overproduction of nNOS and HAO, depletion of 5HT and AchE, upregulation of Bax and downregulation of Bcl-2, increased p53 expression, activation of Caspase-3 and Caspase-9 leading to apoptosis.

**PLATINUM**

Platinum is a rare metal and is considered as very precious. It is one of the members of PGE group elements (PGE) and has led to concern over potential environmental and biological accumulation. PGE group elements include platinum, palladium and rhodium. These are metals that occur at very low concentration in the environment, around 1ng g^-1 in the earth crust and 1pg g^-1 in sea water. PGE are exclusively used in electronics, jewellery production and in pharmaceutical industry. However, there extensive use as vehicle exhaust catalysts (VECs) has resulted in increased release and distribution of these elements in the environment. In nature PGEs occurs primarily as alloys consisting mainly of platinum and are non-reactive and non-toxic. However, use of these elements in various industries particularly as VECs renders them to accumulate in the airborne particles, road dust, soil, mud and water from where they are activated particularly by biotransformation. This allows their entry in organisms and finally bioaccumulation mainly in kidney, liver, spleen and adrenal glands. Human exposure to PGEs occurs mainly through occupational sources.

Platinum compounds, especially the soluble salts are toxic and are responsible for the development of an allergic syndrome known as Platinosis which is characterized by respiratory and cutaneous hypersensitivity on chronic occupational exposure. The symptoms remain from few months to years. Other symptoms of platinum toxicity includes irritation of nose and upper respiratory tract, with sneezing and coughing, and sometimes even asthmatic symptoms such as tightness of the chest, wheezing and shortness of breath. Platinum (II) have also been reported to bind to metallothionein and other proteins depending on the availability of sulfhydryl groups. Platinum based anti-cancerous drug (cispalatin, carboplatin, and oxaliplatin) accounts for intentionally administered in patients. These besides desired effects manifest adverse effects including neurotoxicity. Certain platinum compounds are also known to be cytotoxic and have mutagenic and carcinogenic effects. Halogenated platinum compounds particularly hexachloroplatinic acid (IV), ammonium and potassium hexachloroplatinate (IV), and sodium and potassium tetrachloroplatinate (II) serve as haptens and induces hypersensitivity reactions causing severe allergy leading to rhinitis, conjunctivitis, asthma and urticaria. Various studies have demonstrated that platinum compounds induce oxidative stress which is responsible for the platinum induced renal, cardiac, hepatic and gastric toxicity.

**MANGANESE**

Manganese (Mn), the silvery-white, electronegative metal which resembles iron is hard, brittle, and is readily oxidized. It is the 26th most abundant element in the earths’ crust and 4th mostly used. It exists in 11 oxidation states starting from -3 to +7 of which +2, +3, +4, +6 and +7 are the most common. It is the essential trace element which is required for the growth, development and metabolism of animals, humans and plants in trace amounts, but which when exceeds beyond required concentration leads to severe toxicity. Manganese is naturally present in food, viz nuts, cereals, legumes, fruits, vegetables, grains, and tea. Anthropological sources of manganese include mining industries, burning fossil fuels, which releases Mn into air and water; pesticides which causes release of Mn into soil. Sources of human exposure include occupational, medical and environmental exposures of which occupational exposure forms the main cause of manganese intoxications. Over exposure to airborne Mn through inhalation is seen in miners in manganese dioxide mines, workers in dry-cell battery factories, smelters and welders. Medical sources of exposure are due to its used as contrast agent in medical diagnostics and in patients administered with parental nutrition containing Mn. Manganese is required for a variety of metabolic processes including those involved in skeletal system development, energy metabolism, activation of certain enzymes, functioning of nervous system, functioning of immunological system, reproductive hormone function, and also acts as an antioxidant that protects cells from damage due to free radicals. It plays an essential role in regulation of cellular energy, bone and connective tissue growth and blood clotting. Manganese is an essential component of over thirty-six enzymes that are used for the carbohydrate, protein and fat metabolism. For example, it is a cofactor for glial specific glutamine synthetase, superoxide dismutase, pyruvate carboxylase carboxylase and arginase.

Manganese absorption occurs via ingestion, inhalation, or dermal routes. Of these absorption through gastrointestinal tract is poor. Inhalation of airborne dust particles containing Mn is the major cause of several Mn intoxications. After absorption it is readily distributed to brain, liver by binding to transferrin, gamma globulin and albumin. Manganese crosses blood brain and blood-placental barriers. The metabolism is similar to that of iron. Manganese is mainly excreted through bile (90%) and faeces.

Despite being an essential metal over-exposure to manganese leads to severe neurotoxicity. The toxicity depends on the valence state of the metal, trivalent being more toxic than divalent. Chronic exposure leads to clinical manifestations similar to that of Parkinson’s disease called Parkinsonian syndrome (manganism); the symptoms include headache and insomnia, memory loss, emotional instability, exaggerated tendon reflexes, hyper-myotonia, hand tremor, speech disturbances and festinating gait. Several months before the appearance of manganism symptoms pre-symptoms appear...
known as “manganese madness” which include irritability, emotional lability, illusions and hallucinations are reported.  

Neurotoxicity is due to its interaction with other essential trace elements, including iron, zinc, copper and aluminium. Excess Mn is also reported to be toxic for cardiac muscle cells and tissues by blocking calcium channels. The metal may cause acute liver toxicity by modulating certain enzymes like 3-hydroxy-3-methylglutaryl coenzyme A and cholesterol 7-hydroxylase which are required for cholesterol metabolism and bile production. Manganese exposure has also been documented to decrease fertility and induce developmental abnormalities in foetuses.

Several mechanisms of Mn neurotoxicity have been reported which include the disruption of mitochondrial metabolism, alteration of iron homeostasis, oxidative stress due to alterations in glutamine/glutamate cycling in astrocytes, inflammation, altered glutamate and dopamine (DA) metabolism.

**ALUMINIUM**

Aluminium (Al) is one of the most abundant metals and third most abundant element belonging to group III of the periodic table. It is silvery white, light in weight, soft, malleable, ductile, electropositive metal comprising of 8% of the earth’s crust. It is extensively used and its alloys and compounds are crucial in many industries like transportation and construction facilities, therapeutic drugs, food processing plants, cosmetics, and in household products like cookware and other utensils. Aluminium is exclusively used in the automotive and aerospace industries because of its light weight. Aluminium is ubiquitously found naturally in soil. Acidification of soil and acid rains stimulates the mobility of the metal to aquatic zones and hence to plants irrigated with this water. Also other natural processes which contribute to the mobilization and distribution of this metal in the environment include weathering of rocks. Besides these natural sources, several anthropological activities lead to the release and accumulation of aluminium in the environment mainly air. Wide spread applications of this metal made its exposure to humans inevitable. Aluminium does not play any role in cellular metabolic processes hence exposure to high loads of aluminium results in severe toxicity.

Sources of human exposure to aluminium include occupational, intentional as therapeutic agents containing aluminium like antacids and buffered aspirin and cosmetics. Other non industrial sources of exposure include food and drinking water due to its use in processing, preservation & packaging of food stuffs, and in purification of water. Clinical manifestations of aluminium include neurodegenerative disorders like Alzheimer’s, Parkinson’s disease; neurobehavioural changes like memory loss and vision loss, loss in learning functions. Other symptoms of aluminium poisoning include extreme nervousness, anaemia, headache, and osteoporosis. Workers (or smelter) in aluminium industry show asthma-like symptoms known as “potroom asthma”, affecting the respiratory tract due to aluminium toxicity. However, other agents like hydrogen fluoride and particulate fluoride may also show such symptoms.

Aluminium absorption in the body may be through oral, nasal, and dermal routes. However, absorption by gastrointestinal tract is very limited. Once absorbed, it enters the blood stream, binds to transferrin and citrate, and is distributed to various organs including brain, liver, lungs, kidney, bone etc. Aluminium also crosses blood-brain and blood-placental barriers. It follows metabolic pathways as that of potassium and iron replacing them during entire course of its distribution. Aluminium is excreted mainly through urine.

In biological systems aluminium does not exhibit any redox activity. However, in vitro and in vivo experiments suggest that high Al concentrations cause oxidative stress. Aluminium induces oxidative stress via different mechanisms, mediated by iron leading to the generation of ROS. Aluminium also affects mitochondrial function leading to ROS generation and oxidative damage. Prolong exposure to aluminium even at low levels is reported to cause lipid peroxidation and zwitter ionic lipids like phosphotidylcholine. Deposits of malondialdehyde (MDA), a product of lipid peroxidation in rat brain exposed to aluminium have also been reported. Aluminium also exerts its toxic effect by decreasing the fluidity of the plasma membrane, myelin membrane and synaptosomal membranes thus affecting the neurotransmission and release/uptake of neurotransmitters. Aluminium down regulates neurotransmission by variety of mechanisms including direct inhibition of enzymes responsible for their production and/or utilization of neurotransmitters. Aluminium alters the cell signalling pathways involving the binding of regulatory proteins to poliphosphoinositides in membranes or those involving phosphotidylinositol derived secondary messengers.

Stimulating the pro-inflammatory signals and decreasing the anti-inflammatory molecules like neurotrophils, nerve growth factors, neurotrophic factors derived from brain have also been identified as possible mechanisms for aluminium toxicity.

Clinical manifestations of aluminium toxicity may include gastro-intestinal disturbances, poor calcium metabolism, decreased liver and kidney function, speech disturbances, softening of the bones, and weak aching muscles. Accumulation of aluminium salts in the brain has been implicated in seizures and reduced mental faculties, dizziness, impaired coordination, and loss of balance and energy.

**IRON**

Discovery of iron was an important era in the development of human civilization. It recognized as an essential metal with significant biological and physiological value. Its biological role extends from heme iron group in hemoglobin to iron-sulphur clusters in respiratory chain enzymes of mitochondria which serves as the powerhouse of cell and provides driving force for metabolism and maintenance of cell.

There are various routes of exposure to iron load in humans. Parenteral administration of iron during the process of blood transfusion is one of such mode which may increase intracellular concentration of iron. It is estimated that 500ml of whole blood may contain 200-250 mg of iron. Inhalation of non-industrial iron molecules in form of particulates especially in the subways, and active and passive smoking may cause
systemic exposure to iron.\textsuperscript{174} The most common mode of iron exposure is by ingesting iron containing eatables including all natural foods like vegetables, corn etc. However, excess of iron loading may be result of consuming food items in which iron compounds are added externally this includes flour, corn meal, farina, rice, ready to eat cereals.\textsuperscript{175} Interestingly, iron load can also get increased by consuming citrus fruits rich in ascorbic acid which facilitates absorption of non-heme iron in duodenum. Ingestion of food cooked in iron vessels, as well as consumption of iron-containing vitamins or nutritional supplements may also account in loading of excess of iron in body.

Iron is essential for various cellular activities, and concentrations of iron in cell up to a certain limits are appreciated for cell survival. Conditions when iron concentrations exceed the critical level in the body displays the dark side of iron causing iron-induced toxicity. Excess of iron is responsible for a large numbers of malfunctions and cellular insults including endocrinological, gastrointestinal, infectious, neoplastic, neurodegenerative, obstetric, ophthalmic, orthopedic, pulmonary and vascular diseases. It also contributes in diseases of aging: Alzheimer’s disease, Parkinson’s disease and Atherosclerosis, mortality and pathogenic invasions. Iron toxicity is a result of two different attributes of the metal.

The active redox form of iron (Fe\textsuperscript{2+}) reacts with cellular HzO\textsubscript{2} and is reduced to Fe\textsuperscript{3+} generating oxygen reactive species (ROS) via Fenton redox reaction.\textsuperscript{176}

\[
\text{Fe}^{2+} + \text{H}_{2}\text{O}_{2} \rightarrow \text{Fe}^{3+} + \text{OH}^{-} + \text{OH}^{-}
\]

These generated ROS further causes cellular damages and imbalances including damage to proteins, DNA\textsuperscript{177,178} and causes lipid peroxidation and polysaccharide depolymerization reactions. Another attribute that accounts for iron-induced toxicity is that it serves as a potential growth promoting agent for almost all pathogenic organisms like bacteria, fungi, protozoa\textsuperscript{179} and for all cancerous cells\textsuperscript{180} thus causing cellular tensions.

**MEDICAL COUNTERMEASURES: CHELATION THERAPY**

Exposures to toxic metals, either accidental or chronic in nature are of serious concern to human health. Various factors including age, sex, occupation, lifestyle and economic status play an important role in identifying the susceptibility to the deleterious effects of these metals. Various serious diseases manifestations have been associated with the chronic exposure to toxic metals including cardiovascular defects like hypertension, metabolic disorders like diabetes, cancers, and cognitive defects like Alzheimer’s disease etc.

Chelation therapy has been the mainstay therapy in metal poisoning and related disorders. The term chelate was first applied by Sir Gilbert T. Morgan and H. D. K. Drew in 1920. They suggested the term for the caliper-like groups which function as two associating units and fasten to a central atom so as to produce heterocyclic rings.\textsuperscript{181} Greek work chelate means claw and the process of ring formation is termed chelation. Understanding the concept of chelation promises its application as drug to reduce the body burden of toxic metal. Chelating agents have 1) at least two functional groups with donor atoms capable of combining with a metal and 2) the donor atoms that must be situated as to allow ring formation with metal atom as the closing member. However, for application in the biological system desired properties extend beyond simple chemical behaviour.\textsuperscript{182} For these compounds to be of therapeutic relevance they must i) cross through physiological barriers into compartments where a toxic metal ion is accumulated, ii) form a stable complex with the metal iii) be able to compete with the biological chelator, if required and remove metal from the site and iv) form chelation complex that is non-toxic and easily excretion, from the site of deposition and body.\textsuperscript{183} Chelating agents available for human use may be classified based on structural properties like polyaminocarboxylic acids, chelators with vicinal -SH groups, b-mercapto-alpha-aminoacids, hydroxamic acids,orthohydroxy carboxylic acids or orthodiphenols, and miscellaneous agents. Commonly known chelating agents used in chelation therapy include calcium disodium ethylenediaminetetraacetic acid (CaNa\textsubscript{2}EDTA), 2,3-dimercaptopropanol also known as British Anti-Lewisite (BAL), D-Penicillamine, meso-2, 3-dimercaptosuccinic acid (DMSA), 2, 3-dimercaptopropanesulfonic acid (DMPS), polyaminocarboxylic acids diethlenetriamin pentacetic acid (DTPA), cyclohexanediaminetetraacetic acid (CDTA), hydroxy carboxylic acid sodium catherol 3, 5-disulfonate (Tiron), dithiocarbamates (DDC), desferoxamine (DFO) and deferriprone (L1), etc.\textsuperscript{184}

Calcium disodium ethylenediamine tetraacetic acid (CaNa\textsubscript{2}EDTA) is the most commonly used chelating agent. The agent is termed as universal chelator however is mainly known for application in therapy against lead poisoning. CaNa\textsubscript{2}EDTA is administered intravenously as dextrose or saline infusion due to poor gastric absorption. Its extracellular distribution forms one of its major limitations of only chelating metal circulating extracellularly along with possible metal (lead) redistribution from other tissues to brain. Flora et al.,\textsuperscript{185} recommended CaNa\textsubscript{2}EDTA inappropriate in lead mobilisation test in children owing to their high vulnerability for lead-induced brain toxicity. Other substantial risk associated with CaNa\textsubscript{2}EDTA therapy include renal damage, arrhythmias, tetany, hypocalaemia, hypotension, bone marrow depression, prolonged bleeding time, respiratory arrest, neurotoxicity, other than minor adverse effects like fatigue, headache and fever, etc.\textsuperscript{186}

British Anti-Lewisite (BAL) or 2,3-dimercaptopropanol (Dimercaprol), another one of the oldest chelating agents is indicated in acute soluble arsenic, gold, and mercury poisoning following ingestion, inhalation, or absorption.\textsuperscript{187} Dimercaprol also is reported to complement CaNa\textsubscript{2}EDTA following immediate administration by rapid removal of lead from blood and the central nervous system (CNS) and assisting in mobilization of lead from skeletal stores\textsuperscript{188} compared to CaNa\textsubscript{2}EDTA monotherapy. The combination decreases mortality rate and likelihood of permanent neurologic deficits from lead poisoning along with being less toxic due to lower doses of each being employed. However, BAL is rather considered most toxic chelators available that restrict its
application to few acute poisoning cases. The drug has low therapeutic index and show brain redistribution of metal. Other disadvantages include difficult storage owing to easy oxidation and painful intramuscular mode of administration due to lipophilicity. Minor adverse drug reactions like fever, conjunctivitis, lacrimation, headache, and nausea, etc may be accompanied by serious effects including infection, liver damage, high blood pressure and heart rate, etc. BAL is rapidly absorbed and intracellularly distributed with metabolites excreted in urine.

Safer derivatives of BAL introduced have rather been most successful. Meso 2,3-dimercaptosuccinic acid (DMSA) and sodium 2,3-dimercaptopropane 1-sulfonate (DMPS) are water soluble dithioles with safer drug profiles. The hydrophilic nature of DMSA facilitates good gastrointestinal absorption thus, allowing oral route of administration seen as a distinct advantage over BAL. DMSA and DMPS are FDA (US and Germany respectively) approved drugs against lead and mercury poisoning respectively. DMPS is more effective than DMSA and BAL (28 times) in removing mercury from kidney and arsenic respectively in animal models. However, DMPS is slightly more toxic than DMSA yet both compounds are less toxic to BAL. Both these drugs show predominantly extracellular distribution with DMPS showing some intracellular distribution also. Hydrophilic property of DMSA responsible for its genesis ironically also is responsible for its most important limitation of chelating extracellular metal. Later results in limited application for acute and sub-chronic cases were the metal still resides in the extracellular compartment and has not deposited intracellularly. Thus, esters of DMSA synthesised with the aim to enhance tissue uptake for intracellular chelation were recently introduced. These mono and diesters show higher therapeutic efficacy with monoesters preferred further due to lower toxicity. Monoisoamyl DMSA (MiADMSA), a C5 branched chain alkyl monoester of DMSA with higher lipophilicity compared to DMSA was found highly effective in reducing heavy metal burden from various organs in chronically exposed animals. MiADMSA thus has been identified as a promising drug candidate which is in its developmental phase. Preclinical data available highlights its efficacy and safety in lead and arsenic acute and chronic toxicity in experimental animals. It is also found capable of mobilizing intracellular bound cadmium. The sulfhydryl groups have also been proposed to provide antioxidant functions. Safety of MiADMSA has been well established in preclinical invitro and invivo models with copper depletion as the only prominent reversible side effect. MiADMSA was also reported to abrogate arsenic-induced developmental toxicity in human embryonic stem cell-derived embryo bodies in studying the efficacy of drugs in a comparable manner with animal models. Overloading and possible toxicities from metals that are conventionally known as essential for the body, such as copper and iron has recently gained awareness. D-Penicillamine (DPA) finds its major application in treating patients of Wilson’s disease for removal of excess copper. It is also used to reduce cystine in the urine (cystinuria) and to treat severe rheumatoid arthritis. DPA follows considerable gastrointestinal absorption thus is administered orally and also via intravenous infusion. Its use has rather been limited owing to drug related toxicities and adverse reactions like nephrotoxicity, hypertension, nephritic syndrome, and various autoimmune reactions. Another drug of choice for copper chelation is Tetraethylentetramine or trientine TETA. Drug despite lower efficacy to mobilize copper than DPA, finds its application in patients with DPA intolerance.

Iron chelating agent such as Deferoxamine (DFO) and Deferiprone (L1; CP20; 1,2-dimethyl-3-hydroxyprid-4-one) are prescribed mainly in the treatment of iron overload associated with blood transfusion disorders. Deferoxamine (DFO) is a siderophore secreted by Streptomyces pilosus, a fungus. It entraps iron by complexation to completely cover the metal surface thus preventing iron catalysed free radical reactions. Accidental cases of iron supplement overdose are managed by mechanical removal of tablet from stomach, supportive care along with chelation with DFOA. Although Deferiprone is considered as a suitable alternative to deferoxamine, combination therapy with the two drugs has been recently proposed. The newest iron chelator introduced, deferasirox (4-[3,5-bis-(2-hydroxyphenyl)-1,2,4]triazol-1-yl]benzoic acid) has been approved by FDA that shows high iron specificity, oral availability and safer drug profile.

**LIMITATIONS AND NEWER STRATEGIES**

A brief description above, of the conventionally used chelating agents highlight their major limitations like adverse drug reactions, metal redistribution and essential metal loss. Further, most of the chelating agents considered safe (CaNa₂-EDTA, DMSA, etc) show partial efficacy in case of chronic metal exposure cases by virtue of their inability to cross physiological barriers. Thus, slow accumulation of metal inside the cells characteristic of chronic poisoning gets difficult to address. One such classical example is the failure of DMSA in the clinical trial held at Bangladesh in chronically arsenic exposed patients. Target metal specificity of any chelating agent accounts for another important criteria to avoid essential metal loss. Thus newer therapeutic strategies for management of metal poisoning needs to be defined.

Our group addressed this issue by identifying some newer strategies like combination therapy, relevant nutritional supplementation like essential metals, natural and synthetic antioxidants with chelation benefits with the conventional chelation therapy. Combination therapy may be defined as prescribing more than one, structurally different chelating agent for more efficient and safer removal of metal from body. The two drugs would act through different mechanism chelating metal from different compartments viz. hard and soft tissue (CaNa₂-EDTA and DMSA) or intra- and extra-cellular matrix (DMSA and MiADMSA), thus additive or synergistic effects may be observed. Moreover, such combination therapy for example with a lipophilic and lipophobic (MiADMSA and DMSA) would limit drawbacks like metal redistribution and form a safer regime due to lower doses prescribed. Experimental evidence shows that such strategies result in not only better reduction in metal burden but also more effective...
recovery in biomarkers, neurological defects and molecular markers. Supplanting conventional chelation therapy with antioxidants or essential metals has also been extensively investigated by our group. Antioxidants such as lipoic acid, N-acetyl cysteine, melatonin, gossypin, captopril etc. show promising recoveries in heavy metal toxicity when co-administered with chelating agents. This may be further supported by the fact that oxidative stress has been recognized as the major toxic mechanism for most heavy metals. Similarly, since toxic metals replace essential metals in the body as discussed in the previous sections essential metal deficiency may lead to more potent toxic effects of heavy metals. Further, chelating agents by virtue of non-specificity may also deplete some essential metals worsening the situation. Thus, co-administration of essential metals like iron, calcium and zinc with the chelating agents has been investigated to reveal promising outcomes. Therefore it may be stated that newer therapeutic strategies have shown superior results compared to conventional chelation monotherapy.

### References


