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Clinic and Forensic signs resulting from exposure to toxic elements

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Carolina de Carvalho Peixoto Machado

*Aos meus pais,
pelo amor, compreensão, altruísmo e apoio imensuráveis,
por todos os ensinamentos e valores que me transmitiram e
que pretendo nunca descorar.
Porque sem eles nada disto seria possível,
e se fosse,
nunca teria a mesma cor.*

*À minha segunda família,
dentro e fora desta casa,
pela ajuda, amizade e companheirismo que levarei para a vida,
pelos momentos que guardarei eternamente no coração e
por terem tornado o meu percurso mais leve,
iludindo as Leis da Física.*

*Ao meu avô Firmino e à minha avó Micas,
as pessoas com mais amor, bondade e ternura com que a vida me presenteou,
por me ensinarem que a sabedoria está na simplicidade da Vida,
por viverem os meus sonhos como se fossem os deles,
pela força que sempre me deram para trilhar o meu Caminho,
sendo Luz nos trechos mais sombrios e
por serem Âncora em dias de tempestade.*

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Clinical and Forensic signs resulting from exposure to toxic elements

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Abstract: Heavy metals are natural components of the earth's crust and their properties and toxicity have been recognized for thousands of years. Nowadays, most of the use of heavy metals is related to the industry and presents as a major source of environmental and occupational pollution. This results in several potential exposures coming from natural sources (through contamination), industrial processes, commercial products, among others. The toxicity of most metals accrues from their highly reactive nature resulting in the formation of complexes with intracellular compounds that impair cellular pathways leading to dysfunction and apoptosis. The most commonly involved organ systems include the nervous system, gastrointestinal, hematopoietic, renal and dermatological. This manuscript aims to collect the clinical and forensic signs related to the poisoning by heavy metals such as thallium, lead, copper, mercury, iron, cadmium and bismuth, as well as other elements such as arsenic, selenium and fluor. Furthermore, their main sources of occupational and environmental exposure are highlighted in this review. The importance of rapid recognition is related to the fact that, through a high degree of suspicion, the clinician could start a treatment, even before the toxicological results are available, which can make a huge difference in these patients outcomes.

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Keywords: Heavy metals; intoxication; pathophysiology; signs and symptoms; clinical and forensic diagnosis.

1. Introduction

Acute and chronic poisoning resulting from exposure to heavy metals is uncommon but represents significant morbidity and mortality if unrecognized [1]. Hereupon, clinicians should be aware of the elements that can cause significant harm to patients if not detected and treated quickly. When intoxication is suspected, the diagnosis is based on a careful history of exposure, physical examination, and laboratory to render the best care [1–3].

The metals that establish the greatest threat to human health include lead, cadmium, mercury, and arsenic (arsenic is a metalloid, but it is commonly classified as a heavy metal). Heavy metals are usually defined as those having a density of more than 5 g/cm³, although there is no consensual definition [4].

Although their reactivity is the reason why metals are toxic, it is also essential to vital physiological processes. Examples of essential elements include iron in oxygen transport, zinc in metabolism and manganese and selenium in antioxidant defense. Others, such as

cadmium and bismuth, do not have a biological role in the organism, which makes these metals only have the potential to cause toxicity [3].

Heavy metal poisoning can be accidental or the result of a suicide/homicide attempt. However, most intoxications occur due to environmental and/or occupational exposure. Metals such as lead and mercury, due to their concentrations in the environment, are more likely to result from environmental exposure [5-8]. On the other hand, arsenic and thallium are two elements known for their use in criminal poisonings [1, 9-13]. This reveals the importance of gathering a detailed history of the patient since it can be essential in the diagnosis of a condition that easily raises suspicion of other pathologies, particularly abdominal and neuropsychiatric ones.

This article aims to present an overview of the pathophysiology, clinical presentation, forensic features, and probable sources of exposure to several elements. Some of these characteristic signs may lead to the suspicion of a specific metal, and consequently to the early detection of the underlying intoxication as well as the early formulation of emergent treatment. As Paracelsus noted, "Poison is in everything, and nothing is without poison. The dosage makes it either a poison or a remedy" [14].

2. Materials and Methods

The electronic search took place in PubMed and Google Scholar concerning signs and symptoms, pathophysiology, history and physical examination, diagnostic and forensic signs of intoxication of heavy metals. The keyword "intoxication" was crossed with thallium, arsenic, lead, fluor, copper, selenium, mercury, iron, cadmium, and bismuth as well as their respective forensic and clinical signs. Furthermore, retrieved journal articles, governmental documents and books were reviewed to expand the sources of information. This research was extended to articles written in all languages. Scientific documents, including books, articles, and government documents, were included in this review, making a total of 401 citations.

3. Thallium

Thallium, a toxic heavy metal of the 13th group, was accidentally discovered in 1861 by Sir William Crookes. The before mentioned discovery occurred due to the dust burning from a sulfuric acid industrial plant. This prompted the sighting of a bright green spectral band that quickly vanished [2, 15].

In the past, thallium was mainly used as a rodenticide but as the number of accidental intoxications increased, several countries banned this type of usage. The use of thallium was also discontinued in the treatment of syphilis, gonorrhea, tuberculosis and trichophytosis [16]. Before 1930, scalp ringworm treatments made use of thallium's depilatory properties up until reports of pediatric deaths lead to the clinical abandonment of the mentioned approach [17].

Modern thallium usage includes the manufacturing of green-colored fireworks, imitation jewelry, optical lenses, semiconductors, low-temperature switching devices, and scintillation counters by acting as a chemical catalyst and as a component of artistic paints. The metal's lethal dose is approximately 10–15 mg/kg [16, 17].

On the medicine realm, it is used in the perfusion scintigraphy of the myocardium and in the detection of certain malignant tumors [16]. Given its rapid myocardium distribution and uptake, small nontoxic dosages of radioactive thallium are still used today in cardiac dysfunction spotting [18].

Thallium salts lack taste and odor, while also having the ability to completely dissolve in liquids, being absorbed at a fast speed and evade detection on routine toxicological reports. Such properties make it a perfect candidate for criminal poisonings [9-11]. Nevertheless, such occurrences are rather rare in most Western societies [18].

The poisonings essentially occur from salt ingestion, but there have been reports of dust or fume inhalation from smelting, skin absorption and even overdose from cocaine and heroin consumption [19, 20].

Upon exposure, thallium is rapidly distributed to all body tissues. Association with the Na/K ATPase channel occurs at 10 times the affinity rate of potassium, which results in its binding activity being interrupted. Once thallium's intracellular stores amass, interference with the proper function of several enzymes occurs by binding sulfhydryl groups located on the mitochondrial membrane, which inhibits cellular respiration, disrupts calcium homeostasis and interact with riboflavin and riboflavin-derived cofactors. Additionally, thallium binds to glutathione which inhibits its activation as well as the ability to metabolize heavy metals causing them to over accumulate in the body [21].

Promptly diagnosing thallium poisoning can be complicated and not always direct, mainly due to the variety of non-specific symptoms that tend to occur. Acute polyneuropathies (painful paraesthesia of hands and legs, particularly the soles of the feet, and distal weakness predominantly in the legs), gastrointestinal symptomatology (vomiting, diarrhea, abdominal pain, or obstipation), and later alopecia and other dermatologic signs must imply the consideration of thallium poisoning [22, 23]. Both peripheral neuropathy and gastrointestinal signs are early features of thallium poisoning [24].

The initial dermatological symptoms are not specific of thallium poisoning. Such symptoms include skin desquamation with keratosis on palms and soles (Fig. 1D), acneform or pustular eruptions on the face, angular stomatitis (Fig. 1C), and painful glossitis with tongue tip [25].

The typical triad unfolds by 2-3 weeks [17] with the appearance of dermatological signs such as the loss of scalp hair (Fig. 1A) and the lateral aspects of eyebrows with relative preservation of the eyelashes, pubic hair and axillary hairs [15] and, at the same time, transverse white lunar stripes on nails (Mee's lines or Aldrich-Mee's lines (Fig. 1E)) appear [26].

Acute hair loss seems to occur due to the binding between thallium and the cysteine sulfhydryl groups found in hair while dermatitis, Mees' lines and neuropathy are likely effects of secondary riboflavin deficiency [15, 18, 27, 28].

A significant feature of acute thallium poisoning is the presence of tapered or bayonet hair (Fig. 1B), an abnormal anagen hair with a tapered dystrophic root [29, 30] as well as darkened hair roots when examined under a light-powered microscope. In humans, this occurrence is reported to be detected as early as 4 days after poisoning [31] and illustrates an optical phenomenon which is caused by the accumulation of gaseous inclusions responsible for light diffraction. This finding can be highly point to thallium poisoning diagnostic before the onset of alopecia [29].

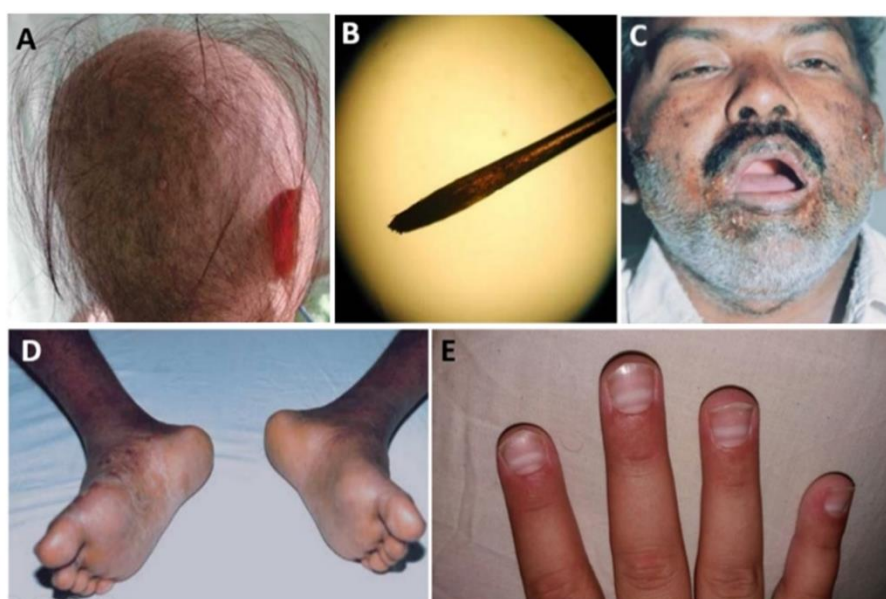


Figure 1. Characteristic scalp hair loss (A), blackened hair root under light microscopy (B), acneform lesions, lip oedema, and angular stomatitis (C), keratosis of soles (D) and Mee's lines (E). Reprinted from (A)- [30] , (B) - [1], (C,D) - [32], (E) -[33].

4. Arsenic

Given arsenic's abundance throughout the earth's crust and soil, it can also be found in rocks, minerals, metals such as lead and copper, as well as all living organisms. Contamination can occur, mainly due to water runoff and leaching [3].

Early reports of its usage date back to 400 B.C. by Greek and Roman physicians [34] and is still used in traditional Chinese and Indian folk medicine [35, 36]. Western societies have recently used it as treatment for late-stage African trypanosomiasis and acute promyelocytic leukemia [3].

With arsenic being prevalent at low background levels throughout our environment its usage is most common on mining operations (for smelting), as herbicide and pesticide for agricultural purposes and in semiconductors and lasers in the electronics industry [37, 38].

Arsenic compounds occur in three oxidation states: elemental, trivalent arsenite and pentavalent arsenate. While elemental is nontoxic, arsenite has ten times more toxicity potential than arsenate. This metalloid occurs in three chemical forms: organic, inorganic and arsine gas. Organic arsenic shows little acute toxicity whereas inorganic arsenic and arsine gas are highly toxic [39].

Even considering that most acute arsenic poisoning occurrences are due to accidental ingestion of arsenic containing pesticides and less commonly from attempted suicide or homicide [12], arsenic is still often seen as the most common homicidal agent. The historical use as a poison has earned it the title of "Poison of Kings and the King of Poisons" [13].

Although inhalation and skin absorption are possible, chronic arsenic exposure mostly occurs by ingestion of geologically contaminated water. There is a natural arsenic occurrence in seafood that has nontoxic organic compounds, such as arsenobetaine, which can lead to higher urine arsenic levels [40, 41]. Metal and electronic worker as well as glass and ceramic manufacturing workers are at the highest risk of occupational arsenic exposure [42].

Binding between inorganic arsenic and the hemoglobin in erythrocytes occurs rapidly after absorption [43]. Redistribution of blood arsenic happens within 24 hours, primarily to the liver, kidneys, heart and lungs. GI tract, spleen and nervous system, are affected in a minor scale. In order to form monomethylarsonic and dimethylarsinic acids which have less acute toxicity, arsenic undergoes hepatic biomethylation [43-46]. Arsenic reaction with the sulfhydryl groups in certain tissue proteins interferes with various enzyme systems vital to cellular metabolism [47]. The lethal dose of inorganic arsenic is approximately 0.6 mg/kg [12].

When acute arsenic toxicity occurs from ingestion, classic gastrointestinal symptoms emerge, including abdominal pain, nausea, emesis, and profuse watery or bloody diarrhea [13, 42, 48]. The mentioned symptoms are followed by hypotension, heart failure, pulmonary edema and shock and can be seen as consequence of cardiomyopathy, ventricular arrhythmias and capillary dilation with fluid loss to third space [13, 42].

Normally, peripheral neuropathy occurs 2 to 8 weeks after exposure, but it can also take place within hours of a severe exposure [13, 49]. The latter consist of a bilateral and symmetric sensorimotor neuropathy, that can easily be misdiagnosed as Guillain-Barré syndrome [13, 50].

When chronic arsenic ingestion takes place, accumulation occurs, predominantly, in the liver, kidneys, heart, and lungs [51]. Even though most of the arsenic in the before mentioned sites is cleared, residual traces remain in the keratin-rich tissues such as nails, hair, and skin. Approximately two weeks after ingestion, arsenic is deposited in the hair and nails. There are several malignancies associated with chronic arsenic exposure, namely in the skin, lung, liver, bladder and kidney [52], with higher prevalence in populations with occupational or environmental exposure to arsenic.

High levels of arsenic, either through ingestion or inhalation results in acute symptoms of Arsenicosis [53]; with Arsenicosis being defined by the WHO working group [54] as a chronic health condition arising from continuous ingestion (not less than 6 months) of arsenic above a safe dosage, usually manifested by characteristic skin lesions, with or without the involvement of internal organs.

Within 1 to 4 weeks, several cutaneous manifestations appear, having been described ranging from raindrop pigmentation - fine freckles of spotted pigmentary changes (Fig. 2H) [55-57] or hypo-pigmented lesions on the trunk and extremities, mild to severe diffused hyperpigmentation or melanosis (Fig. 2D and 2E) [58, 59]. While most intense on the trunk, pigmentation can be localized or diffuse, with a particular effect on skin folds. Macular areas of depigmentation may occasionally appear on normal skin or hyperpigmented background, known as leucomelanosis. Mucous membranes, like the undersurface of the tongue or buccal mucosa, may develop a characteristic pigmentation, the blotchy pigmentation [55-57].

Roughly 5 weeks past the arsenic exposure, the appearance of a transverse white stria, 1-2 mm in width, occurs above the lunula of each fingernail (Mee's lines (Fig. 2C)). This feature is a consequence of the nail matrix interruption, can be seen in acute and chronic poisoning and are not arsenic pathognomonic [60].

The earlier and most common of several cutaneous features is melanosis [61, 62]. Hyperkeratosis of palms (Fig. 2B) and soles (Fig. 2A) is considered to be pathognomonic of chronic Arsenicosis, appearing after prolonged ingestion of arsenic. Therefore, this feature is suggested to be the most sensitive marker for early stage detection of arsenicosis [63]. In a study by *Rattner et al.* [64], for an exposure of 4.75 mg/day, hyperpigmentation appeared after six months and hyperkeratosis occurred 3 years after.

Depending on the extent and severity, keratoses may be classified as mild, moderate, or severe [65]. When it reaches the severe form, it may form keratotic elevations more than 5 mm in size, which are considered as an early marker of carcinogenicity. Exposure to arsenic is related to 3 types of skin cancer, namely, Bowen's disease (Fig. 2F and 2G), basal cell carcinoma, and squamous cell carcinoma [66].



Figure 2. Hyperkeratosis of soles (A) and palms (B), Mee's lines (C), diffused hyperpigmentation (D) and hyperkeratosis (E) on the trunk, Bowen's disease on head (F) and palm (G) and raindrop pigmentation (H). Reprinted from (A,B) - [67], (C) - [68], (D,E,F) - [69], (G) - [66], (H) - [70].

5. Lead

Lead (Pb) is heavy metal with 82 atomic number in the periodic table. Pb in Latin language is called as “Plumbum”, also known as liquid silver, which gave its intoxication the historical name of plumbism or saturnism [71-73].

Several ancient civilizations dedicated themselves to the exploitation of lead, such as the ancient Egyptians, Greeks and Romans. It is speculated that several Roman leaders were victims of lead poisoning and consequently suffered from neurotoxicity and sterility. The beginning of the industrial revolution led to an increase in the consumption of leaded products, mainly gasoline and lead-based paints, whose use became widespread from the beginning of the 20th century. [74].

In England, an outbreak of lead toxicity arose in 1700, caused by lead contaminated cider. The victims developed intense abdominal pain and were said to have “Devonshire Colic” [75]. Benjamin Franklin, who was in the United States, knew the effects of lead intoxication and, in 1763, described abdominal pain and peripheral neuropathy caused by lead [76]. Lead poisoning began to decline progressively in developed countries, due to increased surveillance of industrial and domestic exposure to lead. However, it has not been extinguished [77]. In developing countries, by contrast, lead poisoning is still present, and, on a worldwide scale, continues the most frequent of occupational poisonings [6]. Industrial exposure is primarily via the respiratory tract. Chronic poisoning less commonly results from ingestion [7, 8] although, in pediatric plumbism, pica is usually the common denominator [78-80].

The manufacture of lead batteries, colored paints, lead compounds, and rubber and glass products can create enough lead dust in the air to cause poisoning. In the demolition industry, poisoning by inhalation of lead oxide fumes can occur. Gun club members and instructors show increased lead absorption, due to exposure to lead dust and lead oxide fumes from bullets and explosive charges, respectively. The application of lead-based paints as well as the manufacture of polyvinyl chloride (PVC) plastics is also a form of lead exposure, although final materials are totally safe [81].

Lead is an electropositive metal that presents high attraction for sulfhydryl groups and thus inhibits sulfhydryl-dependent enzymes, particularly intracellular calcium channels. This results in a deficient haem production, proximal renal tubular and osteoblast dysfunction. Among others, lead also affects the vasomotor action of smooth muscle as a result of its effect on Ca-ATPase, which can produce abdominal pain [82].

Lead can deteriorate the integrity of the blood-brain barrier by breaking the intracellular junction of capillary endothelium. This increases the capillary leak into the CNS and, consequently, an increase in the intracranial fluid. This metal can also affect several neurotransmitters, namely, acetylcholine, γ -aminobutyric acid (GABA) and dopamine, causing their spontaneous release. This causes N-methyl D-aspartate (NMDA) glutamate receptors to be blocked and the increase of protein kinase C levels [83, 84].

Any trail of lead in the human body can be seen as contamination since there is no known physiological role. The United States Centers for Disease Control and Prevention established, in 2012, the average level of lead in the blood for adults to 10 $\mu\text{g/dL}$ and for children to 5 $\mu\text{g/dL}$ [85].

When human exposure to lead reaches high levels, damage occurs to almost all organ systems especially the CNS, kidneys and blood, eventually leading to death. At lower levels, heme synthesis, as well as other biological processes, are damaged, psychological and neurobehavioral functions are impaired, and a variety of other effects arise, including headache, memory loss, anemia, abdominal pain, numbness of the extremities, muscle weakness, kidney failure and male reproductive impairment [86]. The main symptoms in young children, where the incidence of lead poisoning is higher than in adults, are irritability, loss of appetite, weight loss, behavioral and learning difficulties, abdominal pain, vomiting, constipation, anemia and renal failure [87].

The standard features to identify lead intoxication involve abdominal pain, anemia with basophilic stippling of red cells, blue-black gum deposits (Fig. 3A e 3B), and a lead line on joint radiography (Fig. 3C) [88]. The blue-purplish lines on the gums are known as ‘Burton's lines’. These are created by a reaction between circulating lead and sulfur ions

released during oral bacterial activity, which leaves lead sulfide at the junction of teeth and gums, creating deposits [89].

In some cases, radiographic imaging helps assist the diagnosis of lead poisoning and also helps to clarify the etiology of exposure. In a patient with potential ingestion of a lead-containing object, an abdominal x-ray is an important exam to be performed as well as in a patient with alleged plumbism or a history of bullet wound, on the region of bullet impact. Children between the ages of 2 and 9 years old, when taking long bone radiographs, may exhibit increased metaphyseal density, also known as “lead lines”. Lead lines are transverse bands of increased density representing bone growth abnormalities, as a consequence of lead exposure. This occurs due to the inhibition of the remodeling of calcified cartilage in the area of provisional calcification under the growth plate – chondral sclerosis. Multiple lines are a sign of repeated exposure to lead [90, 91].

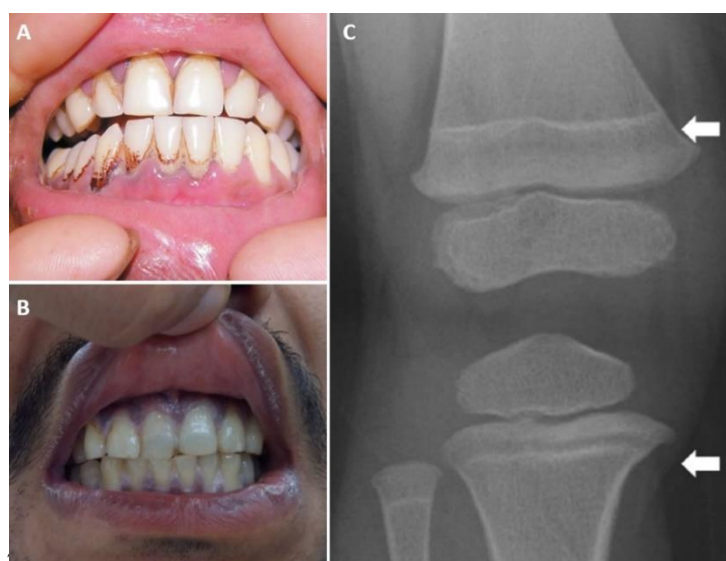


Figure 3. Characteristic Burton's lines are highlighted by the deposits (A) and blue-purplish discoloration of the gums (B). X-ray showing 'lead lines' (C). Reprinted from (A) - [92], (B) - [93], (C) - [94].

6. Fluor

Fluorine (F) is a single highly electronegative ion, non-metal element with 9 atomic number in the periodic table. F is the thirteenth most abundant element and occurs in rock phosphates, minerals and earth's crust in its ionic form. It is essential for normal growth and development of various organs in our body, namely, bone and teeth. For this reason, it is considered to be one of the essential microelements for the organism [95].

Fluor impairs the enamel formation through the reduction of the calcium concentration in the matrix that ultimately affects protease activity, slowing down or blocking the protein degradation in the enamel matrix. This impairment is dependent of the time and dose of exposure [96, 97]. This can result in an interruption of enamel development and a consequent hypomineralization (increased porosity) of the enamel, allowing the appearance of white or yellowish lesions on the surfaces of the teeth [98].

In the skeleton, fluor has a half-life of around 7 years. About 50% of the fluoride becomes incorporated in the hydroxyapatite crystals, by replacing the hydroxide ions, thus altering the structure and size of the crystals. Furthermore, fluoride seems to influence bone turnover through the influence in factors like Runx2 and RANKL [99]. In addition, it affects the expression of osteocalcin and osteoprotegerin and increases osteoblast activity [100, 101].

There are several sources of fluoride and all of them have the capability to produce dental (DF) and skeletal fluorosis (SF) including natural fluoride, artificial or added

fluoride in drinking water and dental products, as much as occupation - related exposures (fluoridated volcanic gases, industrial waste, or the burning of coal fires) [102, 103]

Since 1945, fluoride has been implemented as a supplement in several public drinking water in other to control dental decay [104]. In the last few decades, due its success, the use of fluoride in different forms and concentrations for the prevention of dental caries has been increasing steadily [105]. Water fluoridation was named, by the US Centers for Disease Control, one of the 10 greatest public health achievements of the past century [106].

Nevertheless, as fluoride has become substantially more used in dental products and was included in food sources (via fluoridated water), several sources of fluoride exposure are now related to the mentioned increase in the incidence of dental fluorosis. Dental fluorosis is reported to be the greatest risk of community water fluoridation [107, 108]. Nutrition is equally crucial to controlling serum fluoride level, as ions such as calcium, magnesium and aluminum can decrease fluoride bioavailability. An insufficiency of these ions in food can also increase fluoride uptake [109]. Severe juvenile SF has been reported by some authors who have associated it with inadequate calcium intake in the diet [110].

In some regions of the world such as China, the Middle East, Southern Asia, Americas, Japan and some parts of Africa, dental and skeletal fluorosis are an issue caused by high fluoride levels in underground water. Fluoride can also be present in areas where the soil has a low level of calcium; like regions with high levels of granite or gneiss. In these areas, relatively high fluoride levels are detected in groundwater. When the fluoride level is higher than 1.5 mg/l (1.5 ppm) in drinking water, dental and skeletal fluorosis can happen [111].

Dental fluorosis, first described by Trendley Dean in 1937, is a developmental condition of enamel as a result of excessive absorption and repeated exposure to small doses of fluoride during all stages of tooth development [112, 113]. The gravity of fluorosis observed is multifactorial but is deeply connected with both the amount and timing of fluoride exposure [114, 115].

Mild cases of dental fluorosis are clinically classified by a white opaque appearance of the enamel, as a result of increased subsurface porosity (Fig. 4A). The first change is the appearance of thin white horizontal lines across the covering of the teeth, with white opacities at the recently erupted incisal end. The white lines appear along the 'perikymata', a term relative to horizontal ridges on the surface of the tooth, which correspond to the incremental lines in the enamel denominated as Striae of Retzius [116, 117]. When exposed to higher levels of fluoride, white enamel lines become increasingly defined and thicker (Fig. 4B). Some irregular cloudy sections and thick opaque bands also occur on the involved teeth. The aggravation of dental fluorosis can cause the entire tooth to turn chalky white and lose transparency. Deeper layers of enamel are affected when exposed too long or too high doses of fluoride; the enamel turns less well mineralized [117, 118]. In serious cases, porosity, pitting, and brown areas related to fragile enamel can frequently be noticed on the surface of the tooth (Fig. 4C). Even so, an assertive diagnosis of the disease should only be reached from an adequate anamnesis, and should not be confused with other types of discoloration and dental stains that present a very similar clinical picture, such as the hypomaturation type of Amelogenesis Imperfecta [119].

The upper incisors are especially vulnerable to such environmental modifications. The incisal section of the central incisors may be susceptible to air when there is insufficient lip closure. As a result, the incisal section will become dried out for long periods, and any porosities will consequently be discerned. Also, the incisal edges/cuspal tips are not overlying dentin, so any alteration in pore volume in these sections will reveal itself as clinically different from that of the remaining parts of the teeth. This can lead to the idea that the incisal section is more affected than the remainder of the surface, when in fact they are equally porous [120].

Like dental fluorosis, skeleton fluorosis is caused by high levels of fluoride via ingestion or inhalation, over a chronic period of time. High fluoride accumulation in bones and joints creates an imbalance in the bone mineral metabolism resulting in bone reabsorption and abnormal levels of calcium (Fig. 4D, 4E and 4F) [121-123].

Skeletal fluorosis is often asymptomatic. Nevertheless, as bones and joints become weaker, patients report pain in hands, feet and lower back, muscle weakness, chronic

fatigue and joint stiffness with the movement being difficult and painful [122, 123]. Fusion of vertebrae, kyphosis with limited spinal mobility, flexion contracture of lower extremities and restricted chest wall expansion, may also be observed [122]. In severe cases, progressive weakness of lower limbs, walking difficulty, and sphincter disturbances reveal the onset of radiculomyelopathy [124].

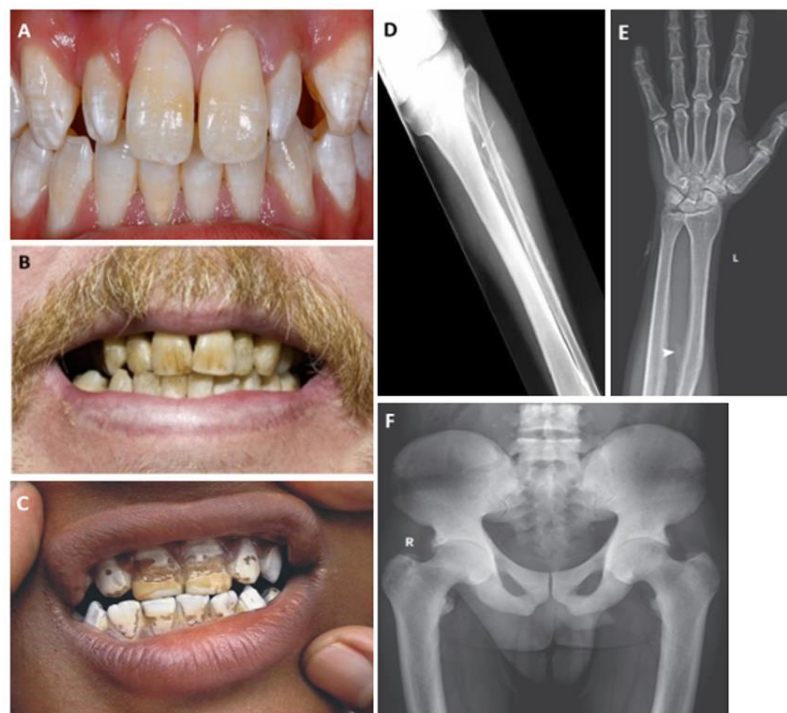


Figure 4. Dental Fluorosis: mild (A), moderate (B) and severe (C). Skeletal Fluorosis: diffuse bone condensation and interosseous membrane calcification of the leg (D) and forearm (E). Pelvic radiography showing osteosclerosis (F). Reprinted from (A) - [125], (B) - [126], (C) - [127], (D) - [124], (E,F) - [128].

7. Copper

Copper (Cu) is the heavy metal of the 11th group with atomic number 29. Copper occurs in nature in the form of different minerals. Of these, the most abundant and economically significant is Chalcopyrite. Copper deposits are classified according to how these deposits were formed. Porphyry deposits (found in mountainous regions of western North and South America) are associated with igneous intrusions and represent two-thirds of the world's existing copper [129, 130]. The type of deposits found in sedimentary rocks (found in areas like the central African copper belt and the Zechstein basin of Eastern Europe) represent about a quarter of copper reserves [131].

It is known that copper was one of the first metals to be manipulated by man. Its use dates back to 8000 B.C., where it was used in ornaments and coins. In 3000 B.C., the discovery of bronze, through the alloy between tin and copper, marked the beginning of the bronze age [132].

Copper's qualities, such as being easily stretched and shaped, being corrosion resistant as well as its ability to conduct heat and electricity, make it a material of choice in a huge variety of domestic, industrial and high-technology applications. Currently, copper is used in civil construction, power generation and transmission, manufacturing of electronic products, production of industrial machinery and transport vehicles, heating and cooling systems, telecommunications, motors, wiring, radiators, touched surfaces (such as brass doorknobs), among others [132]. Furthermore, copper sulfate is commonly

used in farming as a pesticide, in the leather industry, in home-made glue and its burning is a common practice among Buddhists and Hindus [133].

Cu is an indispensable element in the human body, acting as a cofactor for many enzyme pathways. This explains why copper homeostasis is extremely delicate. Copper injury is thought to be derived from the interaction between the reduced form of copper and oxygen, causing the production of toxic oxygen species (TOS) such as superoxide anion, hydrogen peroxide and hydroxyl radicals that have the capacity to be extremely toxic to cells and tissues [134, 135]. The liver can develop acute apoptosis and necrosis due to high concentrations of TOS and low glutathione, which affects the reduction capacity of hepatocytes [136].

Copper-induced (copper toxicity) can be primary, by a metabolic congenital defect, or secondary, when results from high intake, increased absorption or reduced excretion, due to pathological processes [137]. In the secondary causes, poisoning may result from consuming acidic foods cooked in uncoated copper cookware, exposure to excess copper in drinking water, copper salt-containing topical creams for burn treatments, through environmental sources, or in suicide attempts (the lethal dose is 10 to 20g). Children are often attracted to the bright color of the hydrated form of copper sulfate crystals which leads to their intoxication [133].

Acute copper intoxication can present with different symptoms such as metallic taste, nausea, vomiting, abdominal pain, heart failure, hepatic failure, renal failure, intravascular hemolysis and ultimately death [138, 139].

Wilson Disease (WD) is a congenital autosomal disorder of copper metabolism produced by a mutation in the ATP 7B copper transporter gene and has a ratio of around 1/30,000-1/100,000 [140]. The copper-transporting P-type ATPase is encoded by the causative gene, ATP7B [141-143]. Mutations that result in a full absence or nonfunctional of ATP7B protein activity are related to early-onset, typically hepatic, severe Wilson's disease. In comparison, these mutations are rarer [144, 145].

Chronically, excess copper will lead to a progressive deposition in the liver and brain, causing cirrhosis, acute liver failure, and nonspecific neuropsychiatric symptoms, like dysarthria, dysphagia, tremors, ataxia, and inability to concentrate [146].

Copper deposition in the cornea creates the classical Kayser-Fleischer ring (Fig. 5A, 5B, 5C, 5D and 5E). These rings are one of the few diagnostic indicators in clinical medicine. Kayser described the rings for the first time in 1902 and then Fleischer did it in 1909 [147, 148]. In 1949, Gerlach and Rohrschneider finally established the fact that the rings were made of copper [149]. Kayser-Fleischer ring has a golden brown, green or yellow coloration visible on the periphery of the cornea, caused by copper deposition in the Descemet membrane. In general, a slit lamp examination is essential for the confirmation. However, in some situations it is visible to the naked eye. The Kayser-Fleischer ring occurs in more than 99% of patients with neuropsychiatric impairment, but barely in 25-50% of patients with liver disease or a pre-symptomatic state. The ring usually vanishes with treatment and its continuity or return implies a non-controlled copper state [150-152].

In 1922, Siemerling and Oloff described an uncommon ocular manifestation of Wilson's disease known as "sunflower cataract" (Fig. 5F, 5G and 5H). They identified similarities between the lens changes seen in a patient with Wilson's disease and those caused by an intraocular copper-containing foreign body [153]. Although quite rare [154], the sunflower cataract is a feature of WD and was initially referred to as a pathognomonic sign of WD almost 90 years ago [155, 156]. Sunflower cataract incidence is lower than the incidence of K-F rings since sunflower cataracts are seen in the later stages of the disease. As molecular genetic studies are becoming available for the early diagnosis of WD, the occurrences of advanced WD are decreasing nowadays [157].

As Duke-Elder described in 1954, SC consists of a thin, centralized opacification that is placed directly below the anterior capsule and encompasses between one-third and one-half of the anterior lens pole surface area. In all situations, the opacification is central and surrounded by other opacifications disposed in ray-like structures around it. This pattern, with a central disk encircled by petals, looks like a sunflower [154, 158, 159].

The presence of SC turned out to have a limited impact on the patient's visual acuity. This should be highlighted, as classical cataracts frequently have a marked impairment

on visual acuity. SC is not a “true” cataract, as it results from reversible copper deposition below the anterior lens capsule [153, 155, 160-162].

Physicians can be guided toward a WD diagnosis, in the presence of SC or K-F rings. Despite that, due to its occurrence in other medical conditions, like the presence of intra-ocular foreign bodies (IOFB) containing copper, it cannot be considered a pathognomonic ocular sign of WD [154]. Comparing the sunflower cataract caused by copper IOFB and the sunflower cataract in WD, the latter is fainter and occurs in both eyes [163].

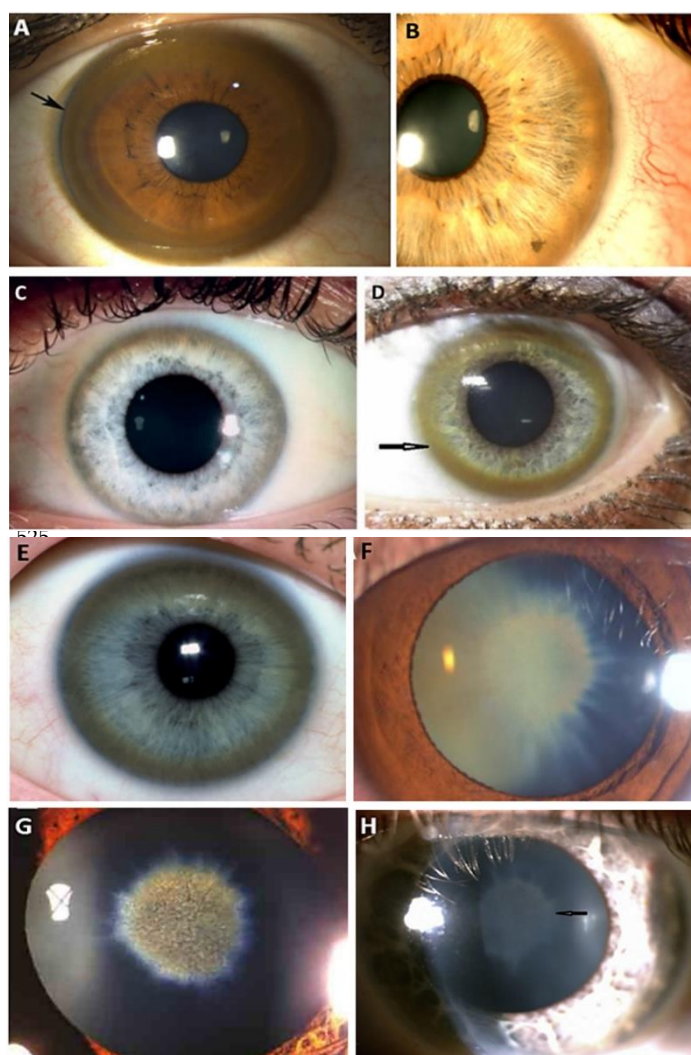


Figure 5. Kayser-Fleischer rings - (A), (B), (C), (D), (E). Sunflower cataracts seen with slit lamp – (F), (G), (H). Reprinted from (A) - [164], (B) - [165], (C) - [166], (D, H) - [151], (E) - [159], (F) - [163], (G) - [167].

8. Selenium

Selenium (Se) is a non-metal element, of the sulfur group, which is fundamental to human and other creatures in residual amounts but is harmful in excess. In 1817, the Swedish chemist Jons Jakob Berzelius recognized, for the first time, the existence of selenium. Nevertheless, its toxicity had been recorded in livestock, for hundreds of years, although the cause was obscure. Within the early 1930s, this ailment known as “alkali disease”, characterized by the loss of hair and hoof, was distinguished as selenium toxicosis [168-170].

Of all the elements, selenium has one of the tightest ranges between dietary insufficiency ($<40\mu\text{g/day}$) and toxic levels (400 mg/day) [171], which makes it vital to carefully control intakes by humans and other animals. The suggested dietary remittance is $55\mu\text{g/d}$ for persons 14 years or older [172, 173]. The sum of selenium accessible in a variable diet with meat, grains, vegetables and nuts is ordinarily adequate to invalidate the need for supplementation [174].

Taking into account that human exposure to selenium is generally subordinate to dietary intakes in food and water, which are essentially controlled by variations within the geology of the Earth's surface, it is critical to understand the relationships between natural exposure and health [175]. Agreeing to the World Health Organization, seleniferous zones have been pointed in South Dakota (USA), Venezuela, and China [176].

Selenium is broadly used in commercial applications, hence being a common source of human exposure. Industrial utilization incorporates solar energy, semi-conductor processing, and the fabricating of electronics and ceramics [177, 178]. It is displayed in steel and copper alloying, used in photographic cells, glass and paint manufacturing, rubber vulcanization, nutritional supplements and shampoos. Furthermore, it is used alongside with different compounds to polish the exterior metallic coating of guns [177, 178]. Selenium salt is widely dispersed in soil, grains and forages [178].

Selenium is a fundamental trace element that constitutes more than two dozen selenoproteins that have a vital part in reproduction, thyroid hormone metabolism, deoxyribonucleic acid synthesis and assurance from oxidative damage and infection [179].

The majority of soluble in water selenium compounds present in nourishment are retained by the gastrointestinal tract (80–95%) [180]. After assimilation, selenium is cleared by the liver and after that carried by selenoprotein P, to all organs, with the highest concentrations happening within the kidney, liver, spleen, testicles and skeletal muscle [181, 182]. The pathways of selenium toxicity in cellular metabolisms are not yet completely cleared. In any case, it is accepted that it can interact with glutathione to form reactive selenotrisulfides and produce harmful species such as superoxide and hydrogen peroxide, oxidizing cell membranes and macromolecules, compromising cell integrity and thus leading to necrosis or cell death [183].

Selenium undergoes an interesting triphasic elimination that depends on its concentration. This element is retained in different organ tissues, and its slow clearance system accounts for the permanence of symptoms [172, 184–187]. Different physical forms of selenium lead to different levels of intoxication, for example, selenide form of selenious acid (monohydrated selenium dioxide, $\text{H}_2\text{O}_3\text{Se}$) has an approximately universal fatal outcome [188].

The emergence of a debate on protective effects of selenium against cardiovascular disease [189] myocardial changes [190] and cancer [191] has driven to expanded utilization of dietary supplements containing selenium in different organic and inorganic forms [192]. Unfortunately, human mistakes related to fabrication resulted in harmful concentrations of selenium in these supplements [193–196].

Occasional cases of acute selenium poisoning are caused by inadvertent ingestion or suicidal attempt [184]. The principal symptoms are intense irritation of the respiratory system and other mucous membranes, metallic taste in the mouth, tingling of the nose and rhinitis. Afterward, edema of the lung and bronchopneumonia can happen. Selenium dioxide (SeO_2) may produce erythema and necrosis of the skin [197].

Comparing acute and chronic poisoning, selenosis resulting from chronic intoxication often results either from consumption of organic compounds in seleniferous plants or livestock, supplements, or by occupational exposure to inorganic selenium in an industrial context [178, 198]. The majority of reported cases of acute selenium poisoning are related to industrial accidents via inhalation of selenium dust and fumes or hydrogen selenide causing irritation of the respiratory tract. The primary symptoms may happen within the first week of exposure and vanish within a few days after the end of the exposure. Treatment can only be symptomatic [178, 197, 199]. Occupational exposure limit for selenium is estimated to be 0.2 mg/m^3 [188].

Selenosis is the classic term for the manifestations of selenium poisoning. Classic signs and symptoms reported are gastrointestinal complaints, metallic taste and a garlic-

like breath odor, alopecia or brittle hair (Fig. 6A and 6B), nail changes – red pigmentation and paronychia (Fig. 6E, 6F and 6G), lack of mental alertness, dermatitis, mucosal irritation, and "rose eye"- a swelling and pink discoloration of the eyelids. The typical garlic odor of breath and sweat is the consequence of the production of dimethyl selenide and reflects high blood concentrations of selenium [192, 197]. More serious presentations include musculoskeletal dysfunction, neuropathy, liver failure, coma and death [178, 197, 198, 200, 201].

The most commonly reported signs, in endemic areas of high concentrations of selenium in the soil, were alopecia and nail dystrophy [202]. Alopecia has been credited to the disruption of structural proteins in keratin; it is formulated that selenium coordinates into disulfide bridges, causing a structural impairment that leads to hair loss [203]. In contrast to androgen-induced hair loss or alopecia areata, induced alopecia by selenium is generalized [204].

The hair becomes dry and brittle and is easily broken off at the scalp, while the follicles remain intact. Usually, hair can be removed by scratching, this results in a rash on the scalp associated with itching. Hair may also fall from the brows, beard, armpit, and pubic area. New hair is always depigmented and loses its luster. The nails become brittle, and white spots and longitudinal streaks appear on the surface. Thumbs seem to be always affected first. The new nail is also dystrophic, and its surface is rough and stripped. Repeated attacks may lead to acropachia (clubbing of the fingers).

Skin lesions have been reported, primarily on the dorsum of hands and feet, lateral side of legs and thighs, the forearms, and the back of the neck. The skin gains a reddish pigmentation (Fig. 6C and 6D), that usually remains, and can develop to being swollen and then blistered and ulcerated. Dental anomalies such as tooth decay and mottling are also observed [202, 205, 206].

The garlic odor and red pigmentation are signs that might lead clinicians to an early diagnosis of selenium poisoning, whether being an inadvertent, suicidal or criminal intoxication [200]. Nail and hair samples may be potentially useful in the study of chronic or past exposure to selenium [178].





Figure 6. Patchy loss of hair over scalp (A), (B). Red-brown discoloration of the vertex scalp (C), (D). Dystrophy of fingernails (E) and toenails (F). Nail dystrophy 43 days after cessation of the exposure (G). Reprinted from (A,E,F) - [206], (B) - [207], (C,D) - [208], (G) - [209].

9. Mercury

Mercury (Hg) is a toxic heavy metal [210] and positions around the 67th place in natural abundance among the elements in crustal rocks. It occurs in the environment in 3 major chemical states: elemental/metallic mercury, inorganic or salts of mercury, and organic or organomercurials (eg, methyl, phenyl, alkyl) [211], which have diverse pharmacokinetic properties [210, 212, 213].

Reported use of mercury dates back to 1500 B.C when the Chinese first mined and used a substance variously known as cinnabar, vermillion, or Chinese red. Around 400 Bc, it is believed that Aristotle called mercury "quick silver," a title that it has held to this day. After that, the Mesopotamians named the planets after metals and Mercury is the only metal that still gives a planet its name [214-216]. The metallic form of mercury has also been named as quicksilver or liquid silver, given its liquid, silvery appearance [217].

For more than 3000 years, mercury and its derivatives have been used as cathartics, antiparasitics, antisypilitics, antipruritics, antiseptics, antiphlogistics, diuretics, vermifuges, dental amalgams, and alteratives [214, 215]. In the 1400s, mercury was used in Western Europe as an antisypilitic which explains the origin of the phrase "two minutes with Venus, two years with mercury" [218].

In 1713, Ramazzini, who is known to be the Father of Industrial Medicine, wrote the renowned treatise "Diseases of Workers," in which he portrayed the toxic manifestations of mercury on surgeons who were treating patients with syphilis by administering mercury inunctions. Although the toxic properties of elemental mercury have been recognized since at least the 1500s, the dependence on mercury during the industrial revolution, resulted in increased occupational and residential exposure [214-216].

Presently, occupational exposure to mercury happens in more than 60 industries, counting those that manufacture glass thermometers, neon lights, paper, paint, jewelry, insecticides, fungicides, batteries, barometers, chlorine, and caustic soda, as well as the occurring within the realm of dentistry [5]. Environmental exposure to mercury occurs, primarily, from the burning of coal by power plants resulting in water pollution and from inappropriate dumping of batteries, paints, lights and industrial products. As the consumption of fish in the human diet increased, as well as the contamination of water and consequently of these animals, environmental intoxication of Hg has become more relevant [219].

By depleting the thiol reserves in the mitochondria and by binding to intracellular sulfhydryl-containing enzymes and proteins, mercury disrupts normal cell biological roles, resulting in apoptosis [220, 221].

Although an overlap in the clinical manifestations of the various forms of mercury is observed, some of these (or combinations of some) are more prevalent in one form of mercury than in another [217, 218, 222, 223]. Therefore, the most commonly reported clinical syndromes of each form of Mercury will be addressed separately.

Elemental

Elemental mercury is a lipophilic, profoundly volatile, heavy, nonwetting liquid that volatilizes to an odorless gas in sufficient amounts to cause clinical toxicity at room temperature [224]. It is known to be one of only two metals that are known to be liquid at room temperature [225]. The vapor pressure of mercury roughly doubles for each 10°C temperature increment, so that its heating significantly increases exposure and toxicity [3]. At room temperature is effortlessly oxidized to mercuric mercury ion when in contact with oxygen [5]. These specific characteristics permitted humans to utilize mercury in a lot of commercial applications such as thermometers, barometers, thermostats, electronics, batteries, dental amalgams, home folk remedies, and several other uses [222, 226].

Toxicity from elemental mercury most commonly emerges from the inhalation of mercury vapors in workplaces exposure through inappropriate dealing of mercury, accidental spills, and poor ventilation. Accidental breakage of mercury-containing devices, like thermometers, can result in residential exposure [5]. The small amount of Hg₀ in a thermometer can cause intoxication in a warm indoor environment [212].

Elemental mercury combines with sulfhydryl groups on cell membranes and interferes with protein and nucleic acid production, calcium homeostasis and protein phosphorylation resulting in oxidative stress and cellular damage [227].

Elemental mercury toxicity occurs from inhalation of the vapor since 80% of inhaled elemental mercury vapor is absorbed and diffuses rapidly across cellular membranes including the blood-brain barrier and placenta [228, 229]. Target organs for elemental mercury vapor include the lungs, brain, and to a lesser degree the kidneys [223, 224, 226]. In the body, elemental mercury has a long half-life of approximately 60 days [228]. Compared to the respiratory tract, GI absorption is insignificant [230] and represents small hazard for patients with intact GI mucosa [231]. In most cases, cutaneous exposure also is of little clinical consequence [224]. Intravenous mercury infusion is an uncommon source of exposure and happens in cases of attempted self-harm [232].

Acute toxicity may manifest within hours of a large exposure with GI complaints, chills, weakness, cough, and dyspnea, with severe cases resulting in adult respiratory distress syndrome [233] and renal failure [234]. While the lung is the critical organ in acute exposure at very high levels, the central nervous system is the main target organ after chronic exposure to mercury vapors [235].

Chronic mercury toxicity may develop over a period of weeks to months, depending on the level of exposure. Primary symptoms commonly mimic a viral illness [223, 236]. Chronic exposure results in two different clinical syndromes, acrodynia and erethism [223, 226, 236].

'Acrodynia', also known as pink disease, Feer syndrome and Feer-Swift disease is an uncommon syndrome that typically emerges from elemental mercury exposure but may also happen in the contact with phenyl mercury and mercury salts [237-239]. In Greek, 'acrodynia' means 'painful extremities' [240]. This intoxication usually occurs in infants and children since they find the shiny, gray appearance of Hg₀ exceptionally appealing [241]. When in contact with elemental mercury through the skin, children can develop acrodynia. Characteristic findings are the following: (1) Autonomic changes – sweating, hypertension, tachycardia; (2) dermatological/dental changes – pruritus, erythematous rash (Fig. 7A and 7B), pink discoloration of nails (Fig. 7C), erythematous gingiva, pink hyperpigmentation on the skin (Fig. 7D and 7E), ulceration of oral mucosa, loose teeth; (3) musculoskeletal – weakness, poor muscle tone [223, 226, 242]. The autonomic changes are caused by the capacity of mercury to inactivate the coenzyme S-adenosylmethionine which inhibits the enzyme catechol-o-methyltransferase, creating elevated levels of catecholamines in the organism. The increment in catecholamines causes hypertension, sweating, and tachycardia that may mirror the presentation of a pheochromocytoma [243-245].

A 24-hour urine sample on these patients uncovers elevated levels of urinary catecholamines, although typically to a lesser degree than that seen in a pheochromocytoma [245].

Patients who have mercury poisoning regularly develop characteristic personality changes collectively named 'erethism'. Neuropsychiatric findings include memory loss, drowsiness, lethargy, depression, withdrawal, irritability, insomnia, shyness, confusion, hallucinations, manic-depressive episodes and emotional lability [226]. Furthermore, incoordination and a fine motor intention tremor primarily involving the hands is also seen in mercury poisonings, accompanied by a rough and rhythmic movement [246, 247]. Later it affects the eyelids, lips, tongue, and head [248, 249]. The nervous system suffers the most prominent damage from the chronic accumulation of mercury within the body and generally results from exposure to air mercury concentrations greater than 0.1 mg/m³ [250].

The classic triad of intention tremors, erethism and gingivitis is the major manifestation of chronic exposure to elemental mercury vapors. Along with the occurrence of gingivitis, hyperpigmentation is often seen and appears as a blue or black line along the gingival margin [251]. Gingivitis and excessive salivation are the complaints most frequently reported [252, 253]. It should be noted that there is a significant clinical overlap between the features of elemental mercury poisoning, especially acrodynia, and those of Kawasaki disease [5].

Inorganic mercury

Inorganic mercury consists of mercuric salts and mercurous salts, the foremost common being mercury(II) sulfide (HgS), moreover commonly known as cinnabar or vermilion [226] and used as a pigment [223]. Mercurial salts have been utilized in a variety of industries including medicine as antiseptics (i.e., mercuric chloride), cosmetics, explosives, dyes and pigments, and as antifungals in paints [223, 226, 254].

The skin is capable of assimilate lethal doses of inorganic mercury and there is a 10% absorption rate through the gastrointestinal tract [238]. Unlike the elemental form, mercury salts are very corrosive to the gut and ingestion is the most common route of poisoning [226, 255]. Inorganic mercury is absorbed promptly through the skin, so patients may develop systemic mercury toxicity even if using only topical preparations [226].

After the ingestion of mercurial salts, patients may complain of oral pain or burning-stomatitis and other gastrointestinal complaints like nausea, vomiting, diarrhea, hematemesis, bloody stools, or abdominal discomfort [226, 255]. Colitis with necrosis or sloughing of the GI mucosa may develop with severe toxicity, since gastrointestinal tract and the kidneys are the target organs of acute inorganic mercury poisoning. [223, 255]. The repetitive cutaneous application can cause hyperpigmentation, swelling, and vesicular or scaly rash. Hyperpigmentation is seen as a gray-brown discoloration being more defined in the skin folds of the face and neck [226].

Like elemental mercury, inorganic mercury can result in acrodynia and erethism [223, 226].

Organic mercury

Although the toxicity of elemental and inorganic mercury has been known for centuries, acknowledgment of organic mercury toxicity has been relatively recent due to several major episodes that have brought it to the forefront of environmental toxicology [3]. Methylmercury, an organic mercuric compound, has resulted in the biggest number of poisonings and is the most prevalent and toxic form of mercury exposure outside of occupational context [3, 5].

In the early twentieth century, organic mercurial compounds were used for industrial and medicinal purposes as preservatives, antiseptics and seed dressings [1].

Nowadays the most common source of organic mercury exposure is dietary consumption of predatory fish through a process known as bioamplification. Inorganic and elemental mercury, from industrial pollution and improper waste disposal, are methylated by soil and marine microorganisms [1, 256]. The methylmercury is rapidly absorbed by plankton algae, which are in turn consumed by fish and other aquatic organisms leading

to higher body concentrations of methylmercury concentrations in the tissues of large predatory fish, such as tuna and swordfish [257].

Due to its solubility, approximately 90% of organic mercury is absorbed in the GI tract [258]. It promptly crosses the blood-brain barrier and placenta reaching levels in the brain three to six times higher than those in the blood [259]. Mothers who were exposed to methylmercury while pregnant, gave birth to children that developed congenital Minamata disease characterized by spasticity, seizures, deafness and severe mental deficiency [256, 260]. The FDA propose an acceptable daily intake of 0.4 µg/kg body weight per day [260].

Organic mercury poisoning is ordinarily chronic and skin signs are thought to be very uncommon. Nevertheless, mucocutaneous hyperpigmentation can appear in chronic absorption and granulomas can develop from mercury injections directly in the skin. Allergic contact dermatitis is the most common form of mercurial reaction in the skin and can occur by both topical and systemic exposure. In acute poisoning, stomatitis can occur and is believed to represent direct irritation of the mucosa [226]. Neurologic signs and symptoms are common but rarely result in the diagnostic since they may mimic other diseases including Parkinson's, Alzheimer's and depression [261].

The most common presentation of organic mercury poisoning is paresthesias of extremities and mouth, incoordination, ataxia, tremor, dysarthria, auditory impairment and concentric constriction of the bilateral visual fields [256, 262]. Autopsy findings commonly report neuronal damage in the gray matter of the cerebral and cerebellar cortex with the most-affected areas being the calcarine region of the occipital lobe and the pre- and post-central and temporal cortex. In the cerebellum there is loss of granule cells typically with preservation of the neighboring Purkinje cells [256, 263]. Damage of peripheral nerves, largely in the sensory fibers, can occur along with the central nervous system impair [263, 264].



Figure 7. Dermatological manifestations of Acrodynia: Rash of lower extremities (A,B), pink discoloration of nails (C), hyperpigmentation with multiple cysts, chloracne-like lesions of face and neck (D) and maculopapular rash in the trunk (E). Reprinted from (A,B,C) - [265] , (D) - [266], (E) - [212].

10. Iron

Iron is the most abundant trace element in the body, an essential element for normal cell metabolism and one of the best-known minerals needed by the human system for homeostasis [267, 268]. This element performs a vital role in metabolic processes such as cellular respiration, myelin formation, development of neuronal dendritic trees, and DNA, RNA and protein synthesis. In addition to this, iron is a co-factor for a plenty of enzymes [269].

The toxicity resulting from intentional or accidental ingestion is a common poisoning. Most of the cases of iron poisoning reported are of pediatric age group, usually in the context of unintentional ingestion [270–272] since iron tablets appear as chocolate candy. Life-threatening toxicity is associated with pediatric ingestion of potent adult preparations, such as prenatal vitamins [273, 274]. The main causes of iron poisoning in adults include suicide attempts and iron overdose during pregnancy [275, 276]. Nowadays, iron intoxication is the second most common overdose in pregnancy [277, 278], with potentially devastating consequences. In addition, iron toxicity may also happen after multiple blood transfusions for the treatment of chronic disorder like thalassemia, sickle cell, and hematological cancers [279].

The serum iron, measured between 2 and 6 hours after exposure, is the most useful laboratory test, since fast iron distribution from circulation into tissues could result in stabilization of the serum iron level at nearly normal limits [270]. When measured at its peak, levels below 350 micrograms/dL, between 350 to 500 micrograms/dL and above 500 micrograms/dL are associated with minimal, moderate and severe systemic toxicity, respectively [279].

The absorption of this metal is carefully regulated to avoid accumulation since there is no physiological mechanism to eliminate iron excess from the body. When iron is bound to transferrin, ferritin, or other transport or storage proteins, it is not available to catalyze the formation of free radicals [280]; in patients with overload iron, the capacity of these proteins to bind with iron is overwhelmed and transferrin becomes saturated. This results in the increase of free iron ion on the serum, which is directly cytotoxic to almost every organ, leading to tissue damage [271, 281].

Iron toxicity is classified as corrosive or cellular. At the cellular level, this element compromises the metabolism in the heart, liver and central nervous system. Free iron enters cells and concentrates in the mitochondria, disrupting oxidative phosphorylation, catalyzing lipid peroxidation, forming free radicals, thus leading to anaerobic metabolism and ultimately leading to cell death [270]. When cellular injury occurs, metabolic acidosis is usual [279]. Its corrosive effect can cause direct caustic damage to the gastrointestinal mucosa, leading to a presentation that may mimic an acute abdomen. The recognition of this clinical constellation as a possible presentation of iron poisoning is imperative. Leukocytosis, hyperglycemia, and metabolic acidosis support the diagnosis of moderate to severe iron poisoning but are nonspecific [270, 282].

The pathophysiology of iron intoxication was initially described by Covey [283] in 1954. This approach has been modified to include five clinical stages. This exact course may not be found in all patients, and in cases of massive overdose, the patient may present in shock. Determination of the iron toxicity stage should be based on symptoms and clinical manifestations and not on the time of ingestion [270, 284]. In stage I, typically occurring between 30 minutes to 6 hours, the patient exhibits gastrointestinal symptoms such as nausea, abdominal pain, vomiting, diarrhea, hematemesis and hematochezias [270, 285, 286]. Stage II is characterized for relative stability and occurs between 6 to 24 hours. It might correspond to true recovery or to the anticipation of clinical deterioration [270, 286, 287]. Circulatory Shock, corresponds to stage III of iron intoxication, emerging typically 6 to 72 hours after the exposure. Patients may present recurrence of GI symptoms, shock, pulmonary dysfunction and metabolic acidosis. Iron-induced coagulopathy, hepatic dysfunction, cardiomyopathy, and renal failure are also observed in this stage [277, 278, 286]. On the IV stage (12 to 96 hours), hepatic necrosis may occur and is characterized by an elevation of aminotransferase levels and possible progression to hepatic failure [277, 278, 286]. Hepatotoxicity is associated with 50% of mortality [288] and it is found to be dose dependent [289]. In the last stage, stage V, the consequences after an acute injury to

the gastrointestinal tract are highlighted. The healing process may include pyloric or proximal bowel scarring and obstruction, with areas of stenosis. These late consequences rarely occur [276-278, 286, 290].

Hereditary hemochromatosis (HH) involves a number of chronic iron overload syndromes characterized by excessive parenchymal iron deposition (Fig. 8B), leading to tissue injury and dysfunction in several organs [291, 292]. It was first described in 1865, by Trousseau [293], when he followed a patient with liver cirrhosis, diabetes mellitus and skin hyperpigmentation, the so called "bronze diabetes". Was only in 1889 that this syndrome was named as "hemochromatosis" (Greek "haima" = blood and "Chromate" = color), by Von Recklinghausen [294, 295].

HH is recognized as the most frequent recessive autosomal disease in European origin population, namely of Celt origin [292, 296, 297]. Approximately 10% of Caucasians in Western countries carry one gene for hemochromatosis [298].

HH is classified in two main groups: HFE mutations and non-HFE mutations [292]. When the HFE mutation is present, this syndrome is known by hemochromatosis type I or classic HH and represents 90% to 95% of all cases of HH [294]. The HFE gene is located on chromosome 6 near the HLA locus, with most of the time a homozygous C282Y mutation [291]. The HFE gene controls intestinal iron absorption and deposition in the tissues, especially the liver, heart, pancreas, pituitary gland, joints, and skin [299].

Non-HFE mutations include HH type II, III and IV. HH type II is the juvenile form (JH), which is related to alteration of the hepcidin protein that regulates the absorption of iron [300]. JH appears in the first three decades of life and has a more severe phenotype than classic HH [301]. HH type III is caused by mutation of the type 2 transferrin receptor gene and type IV to a mutation in the *slc40* gene which encodes the ferroportin gene: a major transporter of cellular iron [302, 303]. Type IV, unlike the other types, has a dominant form of transmission [303].

The symptoms and signs are dependent on the insidious accumulation of iron, which occurs steadily and slowly over decades. Most of the patients are asymptomatic until later in life; men develop signs and symptoms between the 3rd and 5th decade and women after age of 50, due to lactation and the physiological blood losses that occur during menstruation and pregnancy [295, 304-309].

The most common and early symptoms include fatigue (70% to 80%), arthralgia/arthritis (40% to 50%), abdominal pain (20% to 60%), decreased libido or impotence (20% to 50%) and weight loss (10% to 50%). The most frequent clinical symptoms for diagnosis are hepatomegaly (50% to 90%), skin hyperpigmentation (Fig. 8A) - a bronze or slate-grey coloration (30% to 80%), hypogonadism (20% to 50%), arthropathy, splenomegaly, diabetes mellitus, cirrhosis, cardiomyopathy and arrhythmia [295, 304-310].

In advanced stages of the disorder, the dysfunction and tissue injury leads to diabetes mellitus, cirrhosis, congestive heart failure, cardiac arrhythmia, and liver cancer [310].

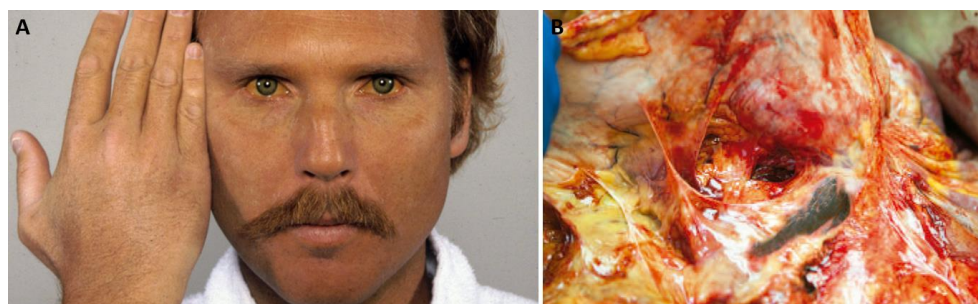


Figure 8. Skin hyperpigmentation- a bronze coloration (A), a black-greyish, rigid pancreas (B). Reprinted from (A)- [311] and (B) - [312].

11. Cadmium

The heavy metal cadmium (Cd) is known to be an ubiquitous trace element and environmental contaminant [313, 314]. This element is one of many metals that are not physiologically or biochemically essential to organisms [315].

Cadmium occurs naturally in ores together with zinc, lead and copper. Although the incidence of chronic cadmium poisoning in industrial workers had declined in the second half of the last century, due to an increased awareness of the effects of chronic exposure to cadmium, its compounds are still widely used in industries [316]. Currently, cadmium is used in the production of anticorrosive agents, as a stabilizer in PVC products, as a color pigment, a neutron absorber in nuclear power plants and, now most commonly, in the fabrication of nickel-cadmium batteries. Cadmium is also present as a pollutant in phosphate fertilizers [316–318].

Cadmium containing products are rarely recycled, which leads us to one of the biggest forms of environmental pollution by cadmium, its dumping and incineration [316]. Compared with mercury or lead, Cd passes more easily from soil to plants and can readily enter the food chain. In addition to this, the uptake process by plants is enhanced at low pH [319].

Its biohazardous potential was first described in 1817, by Friedrich Stromeyer, in those working in zinc smelters. In 1948, emphysema and proteinuria in industrial workers exposed to Cd dust was described by Friberg, linking cadmium exposure to renal toxicity [317].

There are three possible ways of cadmium absorption: pulmonary, gastrointestinal and dermal. Cigarette smoking is a major source of cadmium exposure. The human lung resorbs 40–60% of the cadmium in tobacco smoke [320] and approximately 10% of cadmium oxide is absorbed into the circulation. The concentration of Cd in smokers is 4–5 times higher in blood, and 2–3 times higher in the kidneys, when compared with non-smokers [319]. In non-smokers, food is the most important source of cadmium exposure, from contaminated soil and water [321].

Cd absorbed into the circulation is rapidly taken up by the liver, where it induces the production of Metallothionein [322]. Cadmium-metallothionein (Cd-MT), is freely filtered at the glomerulus and reabsorbed by the proximal tubule. MT bound Cd has a very long half-life, anywhere between 10 and 30 years, and so it accumulates slowly over many years. The MT-Cd complex is a protective response to limit toxicity from free cadmium (Cd^{2+}), but when the production of MT is exhausted, by chronic exposure to Cd, the intracellular levels of Cd^{2+} increase, accumulates in mitochondria and inhibits the respiratory chain by acting at the level of complex III [323]. This leads to the production of reactive oxygen species (ROS) [324], causing progressive tubular cell damage [325–327]. During every person's lifetime, the quantity of Cd in the kidney tubular cells increases [328].

The tissues most affected by chronic Cd toxicity are the kidney, skeleton and lungs. Approximately 50% of the accumulated dose is stored in the kidneys, which makes the kidney the main organ for long-term cadmium accumulation [329, 330]. The primary marker of kidney damage is evidenced by an increased excretion of low molecular weight proteins such as β_2 -microglobulin ($\beta_2\text{M}$) and retinol-binding-protein (RBP) and enzymes like N-acetyl- α -D-glucosaminidase (NAG) [319, 321, 331, 332]. This is called 'tubular proteinuria' and reflects a good index of proximal tubular damage [318]. The urinary concentration of cadmium reflects past exposure, body burden and renal accumulation, while blood concentration serves as a reliable indicator for a recent exposition [333].

The most important metabolic parameter for cadmium uptake is a person's possible deficiency of iron. People with low iron reserves showed a 6% higher uptake of cadmium than those with a normal iron supply [334]. This may explain why children or menstruating women tend to have higher Cd levels [335] when compared with men who have experienced similar levels of environmental exposure. Low iron blood levels stimulate the expression of DCT-1, a metal ion transporter in the GI tract, serving as a gate for cadmium resorption [336].

Acute intoxication is now rare in Europe and North America and its symptoms depend on the route of ingestion. Acute exposure to Cd fumes leads to shortness of breath,

lung edema and destruction of mucous membranes as part of cadmium-induced pneumonitis [337, 338].

The intake of cadmium-contaminated food causes desquamation of the intestinal mucosa, resulting in severe and bloody diarrhea and vomiting [339, 340].

The association between cadmium and skeletal damage was first described in 1950, in Japan, where the exposure was caused by cadmium contaminated water used for irrigation of local fields [319, 341]. This disease became known as Itai-Itai [342] and the affected population showed a wide range of symptoms such as low grade of bone mineralization, high rate of fractures, increased rate of osteoporosis, and intense bone pain (a combination of osteomalacia and osteoporosis) [343].

In the 20th Century, sundry studies showed that exposure to cadmium-polluted fume and dust, e.g. in industry workers, also led to the development of bone damage [344]. Furthermore, during recent years, new data suggests that relatively low cadmium exposure, is enough to give rise to skeletal damage, evidenced by osteoporosis and fractures [345-347].

In addition to the effects on the kidney and bones, chronic Cd toxicity causes infertility [348] and, more recently, has been linked as a potential cardiovascular risk factor, shown by an increased risk of myocardial infarction [349], development of hypertension [350] and diabetes mellitus type 2 [351].

12. Bismuth

Bismuth (Bi) is a trivalent poor metal that chemically resembles arsenic and antimony [352, 353]. Is thought that its designation derives from the German word "wismuth" (white mass). It is the least abundant element of the 15th group and a relative rare element. Bismuth is naturally monoisotopic (²⁰⁹Bi) and is considered to be the heaviest stable element since its theoretical half-life is 1.9×10^9 years. It is sourced as a by-product of lead, copper and tin mining, being primarily found as bismuthinite (bismuth sulfide) and bismite (bismuth oxide) ores [354].

Until today, no natural biological role of this metal was found. There are reference to inorganic and organic bismuth compounds uses in medicine dating back to the late 18th century [355]. They have been used in a variety of diseases such as skin conditions and treatment of war wounds, through topical application (as astringents, emollients and antimicrobials), syphilis by parenteral administration and orally to cholera infantum, peptic ulcer and other gastrointestinal disorders [356-359].

Although the demand for these components declined dramatically after the appearance of antibiotics, for example in the treatment of syphilis, in 1980s, with the discovery of *H.pylori*, bismuth emerged again as an effective treatment for many gastrointestinal complaints [360].

Pepto-Bismol (bismuth subsalicylate, BSS) is a preparation developed in 1901 and still currently used in the treatment of gastritis and dyspepsia. De-Nol (colloidal bismuth subcitrate, CBS) and Pylorid (ranitidine bismuth citrate, RBC) are other bismuth medications, used currently in the treatment of ulcers and *H. pylori* infection [352, 353, 361].

Popularized in the First World War, Bismuth iodoform paraffin paste (BIPP) gauze is an antiseptic dressing [362], still used frequently by maxillofacial surgeons and otolaryngologists. Furthermore, BIPP is used in the management of epistaxis [363]. However, neuropsychiatric symptoms can develop from the use of BIPP gauze. In order to prevent it, clinicians should take in account the patient's renal and hepatic function as well as the size and condition of the wound [364].

Absorption is primarily via the gastrointestinal tract [365] and although organic bismuth compounds show minimal absorption (for CBS 0,5% of the dose), it is still significant. Inorganic compounds with poor solubility are not absorbed [366-368]. Solubility is increased in the presence of hydroxy and sulfhydryl groups and by the acidity of the medium. Moreover, seems like the absorption of bismuth is aim of high inter and intraindividual variation, for example, through the presence of intestinal microorganisms that may convert bismuth to its soluble form and contributes to its toxicity [369-371].

After bismuth enters the bloodstream, Bi is distributed in all tissues with high concentrations in the kidneys and liver [365]; the target organs of acute CBS toxicity. Independent of the bismuth compound administered and the route of administration, its concentration and retention time in the kidney is always higher than in other organs (eg, lung, liver, brain). Bismuth binds to a metal binding protein in proximal renal tubule cells and remains bound for months. Its toxicity is attributed to its affinity to combine with sulfhydryl groups of vital enzymes leading to destroying the functions of these enzymes [366, 369]. Since greater than 99% of ingested bismuth is not absorbed, it makes it suitable as a locally acting gastrointestinal agent [372]. The residual compounds that are absorbed are eliminated unmetabolized by the urinary and hepatobiliary routes. The exact proportion contributed by each route is still a subject of discussion [369, 373].

Serum and urine levels of bismuth, in people who are not in contact with bismuth compounds, are between 1 and 15 µg/L (+/- 12) and less than 1 µg/L, respectively [366, 369]. Taking therapeutic bismuth compounds can lead to serum levels between 10-20 µg/L and serum levels greater than 50 µg/L can be a sign of toxicity [366].

The reported toxic effects attributed to bismuth poisoning include hepatitis and hepatic fatty degeneration [374], nephropathy [375], osteoarthritis [369], gingivostomatitis [372], colitis [366] and several neuropsychiatric symptoms [365, 372, 376, 377].

Generally, acute bismuth toxicity (single high dose) manifests as nephrotoxicity, whereas chronic exposure to high levels of bismuth salts result in encephalopathy [352, 366]. Severe overdose of acute bismuth induces reversible proximal tubular damage leading to acute tubular necrosis. Acute tubular necrosis results in proximal tubular dysfunction (Fanconi's syndrome), which clinically presents by hypophosphatemia, hypouricemia, metabolic acidosis, renal glycosuria and tubular proteinuria. Bismuth is known to cause a reversible Fanconi's syndrome and to following the path of acute tubular necrosis [378, 379]. Several heavy metals intoxications are described to cause reversible Fanconi's syndrome such as bismuth, lead and mercury [380], unlike chronic cadmium intoxication which has been linked with irreversible Fanconi's syndrome [381].

Bismuth crosses the blood-brain barrier and bind to enzymes involved in oxidative metabolism, resulting in reduced oxygen consumption and cerebral perfusion. This can lead to neuropsychiatric symptoms and encephalitis with brain lesions [366, 382]. There were several reports of neurotoxicity, from the use of BIPP impregnated gauze, since a well-study outbreak of bismuth encephalopathy, in 1970, established the typical symptoms of oral bismuth toxicity to be depression, anxiety, irritability, and possibly mild incoordination [371]. Through the years, several clinicians reported more neuropsychiatric symptoms developed after procedures with the use of BIPP gauze: Malaise, insomnia, personality change [362], ataxia [383], dysphonia [384], dysarthria [385], gait dyspraxia [384], myoclonic jerks [386] - more common in the distal upper limbs [369], delirium, drowsiness [386] and coma [371].

Another characteristic feature of bismuth ingestion is the gingivostomatitis with bluish black gum discoloration, gingivitis and ulceration [387-391]. Chronic intoxication from repeated oral or parenteral doses causes "bismuth line"; a gum condition with black spots of buccal and colonic mucosa [392]. Blackening of the tongue and teeth (Fig. 9) has been reported as a harmless side effect of bismuth toxicity [393]. It has been suggested that the oral lesions may be secondary to the nephrotoxic effect of the bismuth compounds [389, 390].



Figure 9. Greyish discoloration of teeth. Reprinted from [394].

13. Conclusions and future perspectives

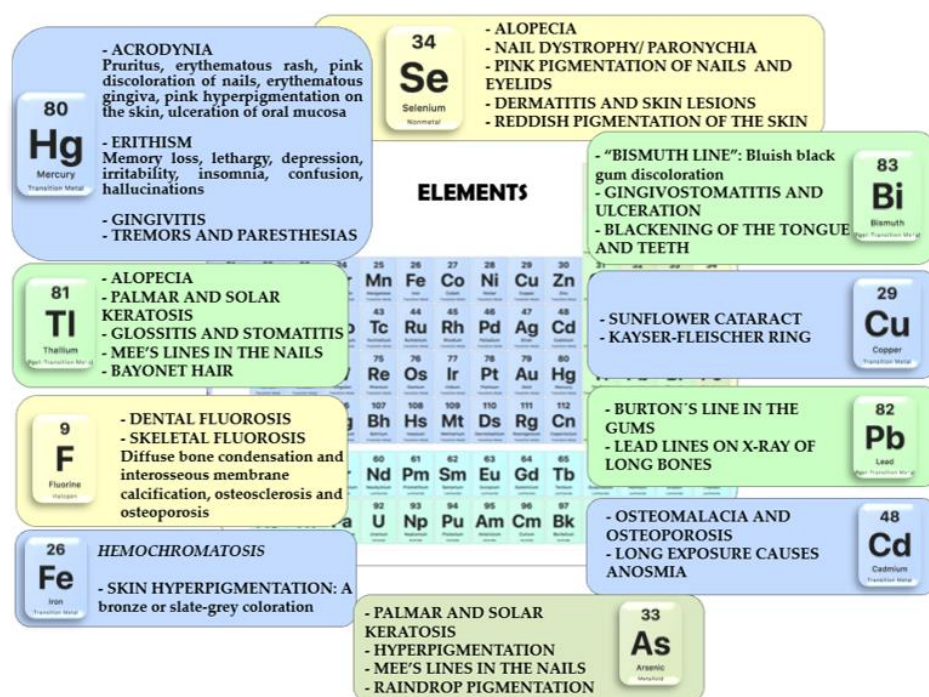


Figure 10. Signs and symptoms associated with poisoning by the addressed elements.

Heavy metals are dense, naturally occurring elements that accumulate in the environment, mostly due to anthropogenic activities, causing health hazards for humans and other creatures [395]. The development of the industry and expansion of the chemical compounds is leading to the increase of the toxic substances emitted to the environment [396]. Environmental pollution can result in contamination of air, water, sewage, seawater, waterways, and can accumulate in plants, crops, seafood, and meat and indirectly affect humans [395]. Some occupations have increased risk for exposure and toxicity of a particular metal such as metal finishing industry workers and traditional glassworks [397]. Toxicity from heavy metals has traditionally been thought to occur only in an industrial or occupational context and to present with severe clinical signs and symptoms, however, nowadays, it is known that poisoning of heavy metals can occur in several ways, with minimal symptoms [318]. The clinical course of intoxication is determined by the element involved, the level and mode of exposure, the chemical and valance states of the compound (elemental vs. organic vs.inorganic), and by the intraindividual variation of the patient [1, 397].

Nevertheless, heavy metal poisoning is still underestimated in the worldwide community. Humans can be exposed through inhalation, ingestion, or contact with the skin. Mining, fossil-burning, agriculture, seafood, consuming plants and meat, that accumulated these metals from contaminated water and soil, are the most common natural causes of toxicity. The incidence and magnitude of the toxicities are dependent on the natural soil content, geographical location, location and number of industries, regulatory measures to contain pollution, healthcare facilities, and individual factors like genetics and nutritional status [398]. Although accidental exposure is far more common, intentional poisoning has been reported in suicide and homicide attempts [1, 9-13].

Acute toxicity results from exposure to large doses, in a short time exposure, and its presentation is usually dramatic. The most common presentations vary with the form of exposure: respiratory symptoms in cases of inhalation, skin lesions in topical contamination and, in cases of ingestion, typically mimic acute gastroenteritis.

In contrast, chronic poisoning occurs due to repeated small amounts resulting in cumulative toxicity. This type of toxicity is usually more difficult to detect, needing a higher level of suspicion. Since chronic poisoning relies on cumulative exposure, scrupulous analysis of the patient history and routine is essential seeing that the recognition of the metal in question is facilitated if the source is known. Bernardino Ramazzini (1633-1714), known as the father of occupational medicine, formulated an injunction that remains the key to healthy occupational practice: ‘what is your occupation?’ [4, 81].

In general, diagnosis of heavy metal poisoning includes the history of exposure, physical findings and samples of blood, urine, skin, nails, hair and gastric content. Of these, the urine sample is typically the most useful for qualitative detection [3, 398, 399]. Management includes the removal of the source and the offending agent (sometimes using chelating agents), supportive therapy and prevention of further exposure [1, 3, 398, 400].

When unrecognized, acute and chronic toxicity from exposure to heavy metals pose significant morbidity and mortality, since treatment is delayed [1]. Hereupon, preventive measures, principally in industrial workers and others at higher risk, such as monitoring air, water, food, screenings of blood and urine for toxicity from heavy metals, should be realized periodically by Health authorities [401]. Furthermore, in order to prevent or minimize exposure of populations, cooperation with health and civic authorities must exist and industries must be sensitized for the existence of a social responsibility with regard to proper wastes disposal, which is a strong source of contamination for World society in general [398].

Compliance with ethical standards

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References

1. Ibrahim, D., et al., *Heavy metal poisoning: clinical presentations and pathophysiology*. Clinics in laboratory medicine, 2006. **26**(1): p. 67-97.
2. Wiley IL, J.F., *Difficult diagnoses in toxicology: poisons not detected by the comprehensive drug screen*. Pediatric clinics of North America, 1991. **38**(3): p. 725-737.
3. Rusyniak, D.E., et al., *Heavy metal poisoning: management of intoxication and antidotes*. Exs, 2010. **100**: p. 365-96.
4. Kuruvilla, A., et al., *Clinical manifestations of lead workers of Mangalore, India*. Toxicology and industrial health, 2006. **22**(9): p. 405-413.
5. Ozuah, P.O., *Mercury poisoning*. Current problems in pediatrics, 2000. **30**(3): p. 91-99.
6. Koh, D. and C.K. Seng, *Textbook of occupational medicine practice*. 2001: World Scientific.
7. Gosselin, R.E., et al., *Clinical toxicology of commercial products*. Vol. 1085. 1984: Williams & Wilkins Baltimore.
8. Lohiya, G.S. and S. Lohiya, *Lead poisoning in a radiator repairer*. Western journal of medicine, 1995. **162**(2): p. 160.
9. Desenclos, J.C., et al., *Thallium poisoning: an outbreak in Florida, 1988*. South Med J, 1992. **85**(12): p. 1203-6.
10. Peter, A.L. and T. Viraraghavan, *Thallium: a review of public health and environmental concerns*. Environ Int, 2005. **31**(4): p. 493-501.
11. Sharquie, K.E., et al., *Outbreak of thallium poisoning among Iraqi patients*. Journal of the Saudi Society of Dermatology & Dermatologic Surgery, 2011. **15**(1): p. 29-32.
12. Sullivan, J.B.K.G.R., *Clinical environmental health and toxic exposures*. 2001.
13. Vahidnia, A., G.B. van der Voet, and F.A. de Wolff, *Arsenic neurotoxicity--a review*. Hum Exp Toxicol, 2007. **26**(10): p. 823-32.
14. Sá, M.I., et al., *Acute iron intoxication in pregnancy: case report Intoxicação aguda por ferro na gravidez: a propósito de um caso clínico*. Acta Obstet Ginecol Port, 2015. **9**(5): p. 417-420.
15. Galván-Arzate, S. and A. Santamaría, *Thallium toxicity*. Toxicol Lett, 1998. **99**(1): p. 1-13.
16. Nelson, L.S., et al., *PESTICIDES*, in *Goldfrank's Toxicologic Emergencies*, 11e. 2019, McGraw-Hill Education: New York, NY.
17. *Thallium Poisoning: Diagnosis May Be Elusive But Alopecia Is The Clue*. BMJ: British Medical Journal, 1993. **306**(6891): p. 1527-1529.
18. Rusyniak, D.E., R.B. Furbee, and M.A. Kirk, *Thallium and arsenic poisoning in a small midwestern town*. Ann Emerg Med, 2002. **39**(3): p. 307-11.
19. Baldwin, D.R. and W.J. Marshall, *Heavy metal poisoning and its laboratory investigation*. Ann Clin Biochem, 1999. **36** (Pt 3): p. 267-300.
20. Yumoto, T., et al., *A Successfully Treated Case of Criminal Thallium Poisoning*. Journal of clinical and diagnostic research : JCDR, 2017. **11**(4): p. OD01-OD02.
21. Yang, G., et al., *Hair Loss: Evidence to Thallium Poisoning*. Case Reports in Emergency Medicine, 2018. **2018**: p. 1313096.
22. Pelclová, D., et al., *Two-year follow-up of two patients after severe thallium intoxication*. Hum Exp Toxicol, 2009. **28**(5): p. 263-72.

23. Rodriguez, J.J., G. Mosqueda-Tapia, and M. Altamirano-Lozano, *Genotoxicity Assessment of Human Peripheral Lymphocytes Induced by Thallium(I) and Thallium(III)*. *Toxicological & Environmental Chemistry*, 2017. **99**: p. 01-28. 1256
1257
1258
24. Dumitru, D. and A. Kalantri, *Electrophysiologic investigation of thallium poisoning*. *Muscle Nerve*, 1990. **13**(5): p. 433-7. 1259
1260
25. Wang, T.-T., et al., *Early diagnosis, treatment, and outcomes of five patients with acute thallium poisoning*. *World journal of clinical cases*, 2021. **9**(19): p. 5082-5091. 1261
1262
26. Liu, C.-H., C.-Y. Huang, and C.-C. Huang, *Occupational Neurotoxic Diseases in Taiwan*. *Safety and Health at Work*, 2012. **3**(4): p. 257-267. 1263
1264
27. Cavanagh, J.B., *What have we learnt from Graham Frederick Young? Reflections on the mechanism of thallium neurotoxicity*. *Neuropathol Appl Neurobiol*, 1991. **17**(1): p. 3-9. 1265
1266
28. Mulkey, J.P. and F.W. Oehme, *A review of thallium toxicity*. *Vet Hum Toxicol*, 1993. **35**(5): p. 445-53. 1267
29. Feldman, J. and D.R. Levisohn, *Acute alopecia: clue to thallium toxicity*. *Pediatr Dermatol*, 1993. **10**(1): p. 29-31. 1268
1269
30. Lu, C.-I., et al., *Short-term thallium intoxication: dermatological findings correlated with thallium concentration*. *Archives of dermatology*, 2007. **143**(1): p. 93-98. 1270
1271
31. Moeschlin, S., *Thallium poisoning*. *Clin Toxicol*, 1980. **17**(1): p. 133-46. 1272
32. Misra, U., et al., *Thallium poisoning: emphasis on early diagnosis and response to haemodialysis*. *Postgraduate medical journal*, 2003. **79**(928): p. 103-105. 1273
1274
33. Almassri, I. and M. Sekkarie, *Cases of thallium intoxication in Syria: a diagnostic and a therapeutic challenge*. *Avicenna journal of medicine*, 2018. **8**(03): p. 78-81. 1275
1276
34. Hunt, E., et al., *Arsenic poisoning seen at Duke Hospital, 1965-1998*. *N C Med J*, 1999. **60**(2): p. 70-4. 1277
35. Lynch, E. and R. Braithwaite, *A review of the clinical and toxicological aspects of 'traditional' (herbal) medicines adulterated with heavy metals*. *Expert Opin Drug Saf*, 2005. **4**(4): p. 769-78. 1278
1279
36. Wong, S.T., H.L. Chan, and S.K. Teo, *The spectrum of cutaneous and internal malignancies in chronic arsenic toxicity*. *Singapore Med J*, 1998. **39**(4): p. 171-3. 1280
1281
37. Hemond, H.F. and H.M. Solo-Gabriele, *Children's exposure to arsenic from CCA-treated wooden decks and playground structures*. *Risk Anal*, 2004. **24**(1): p. 51-64. 1282
1283
38. Wester, R.C., et al., *In Vivo Percutaneous Absorption of Arsenic from Water and CCA-Treated Wood Residue*. *Toxicological Sciences*, 2004. **79**(2): p. 287-295. 1284
1285
39. Gorby, M.S., *Arsenic poisoning*. *The Western journal of medicine*, 1988. **149**(3): p. 308-315. 1286
40. Lai, V.W., et al., *Arsenic speciation in human urine: are we all the same?* *Toxicol Appl Pharmacol*, 2004. **198**(3): p. 297-306. 1287
1288
41. Le, X.C., et al., *Speciation of key arsenic metabolic intermediates in human urine*. *Anal Chem*, 2000. **72**(21): p. 5172-7. 1289
1290
42. Department of health and human services, U.S.A., *Toxicological profile for arsenic*. *Agency for Toxic Substances and Disease Registry*, 2007. 1291
1292
43. Winski, S.L. and D.E. Carter, *Arsenate toxicity in human erythrocytes: characterization of morphologic changes and determination of the mechanism of damage*. *J Toxicol Environ Health A*, 1998. **53**(5): p. 345-55. 1293
1294
1295
44. Carter, D.E., H.V. Aposhian, and A.J. Gandolfi, *The metabolism of inorganic arsenic oxides, gallium arsenide, and arsine: a toxicochemical review*. *Toxicol Appl Pharmacol*, 2003. **193**(3): p. 309-34. 1296
1297

45. Mahieu, P., et al., *The metabolism of arsenic in humans acutely intoxicated by As₂O₃. Its significance for the duration of BAL therapy.* Clin Toxicol, 1981. **18**(9): p. 1067-75. 1298
1299
46. Radabaugh, T.R. and H.V. Aposhian, *Enzymatic reduction of arsenic compounds in mammalian systems: reduction of arsenate to arsenite by human liver arsenate reductase.* Chem Res Toxicol, 2000. **13**(1): p. 26-30. 1300
1301
1302
47. Hu, H., *Heavy Metal Poisoning*, in *Harrison's Principles of Internal Medicine*, 20e, J.L. Jameson, et al., Editors. 2018, McGraw-Hill Education: New York, NY. 1303
1304
48. Vantroyen, B., et al., *Survival after a lethal dose of arsenic trioxide.* J Toxicol Clin Toxicol, 2004. **42**(6): p. 889-95. 1305
1306
49. Kishi, Y., et al., *An epidemic of arsenic neuropathy from a spiked curry.* Neurology, 2001. **56**(10): p. 1417-8. 1307
1308
50. Donofrio, P.D., et al., *Acute arsenic intoxication presenting as Guillain-Barré-like syndrome.* Muscle Nerve, 1987. **10**(2): p. 114-20. 1309
1310
51. Benramdane, L., et al., *Arsenic speciation in human organs following fatal arsenic trioxide poisoning—a case report.* Clin Chem, 1999. **45**(2): p. 301-6. 1311
1312
52. Jackson, R. and J.W. Grainge, *Arsenic and cancer.* Can Med Assoc J, 1975. **113**(5): p. 396-401. 1313
53. Caussy, D., *A field guide: detection, management and surveillance of arsenicosis in South-East Asia Region.* 2006, New Delhi: WHO Regional Office for South-East Asia. 1314
1315
54. *Arsenicosis Case-Detection, Management and Surveillance. Report of a Regional Consultation.* Who regional Office for South-East Asia, 2003. 1316
1317
55. Chilvers, D. and P. Peterson, *Global cycling of arsenic. Lead, mercury, cadmium and arsenic in the environment*, 1987. **31**: p. 279-301. 1318
1319
56. Coles, D.G., et al., *Chemical studies of stack fly ash from a coal-fired power plant.* Environmental Science & Technology, 1979. **13**(4): p. 455-459. 1320
1321
57. Kapaj, S., et al., *Human health effects from chronic arsenic poisoning—a review.* Journal of Environmental Science and Health, Part A, 2006. **41**(10): p. 2399-2428. 1322
1323
58. Cheng, C.N. and D.D. Focht, *Production of arsine and methylarsines in soil and in culture.* Appl Environ Microbiol, 1979. **38**(3): p. 494-8. 1324
1325
59. Nakamura M, M.Y., Tanaka S, and H. Y, *Chemical form of arsenic compounds and distribution of their concentrations in the atmosphere.*, ed. A.O. Chem. 1990. 4:223-30. 1326
1327
60. Mees, R., *Een verschijnsel bij polyneuritis arsenicosa.* Verloskd Gynaecol. Ned Tijdschr Verloskd Gynaecol, 1919: p. 1:391–6. 1328
1329
61. Milton, A.H., et al., *Non-cancer effects of chronic arsenicosis in Bangladesh: preliminary results.* Journal of Environmental Science and Health, Part A, 2003. **38**(1): p. 301-305. 1330
1331
62. Saha, K.C., *Diagnosis of arsenicosis.* J Environ Sci Health A Tox Hazard Subst Environ Eng, 2003. **38**(1): p. 255-72. 1332
1333
63. Kadono, T., et al., *Skin manifestations of arsenicosis in two villages in Bangladesh.* International Journal of Dermatology, 2002. **41**(12): p. 841-846. 1334
1335
64. Rattner, H. and M. Dorne, *Arsenical pigmentation and keratoses.* Arch Dermatol Syphilol, 1943. **48**: p. 458-60. 1336
1337
65. Ghosh, P., et al., *Epidemiology and prevention of chronic arsenicosis: an Indian perspective.* Indian J Dermatol Venereol Leprol, 2008. **74**(6): p. 582-93. 1338
1339

66. Das, S., J. Chowdhury, and L. Ghoshal, *An introspection into the cutaneous manifestations of chronic arsenicosis as reported in a tertiary care centre in Kolkata*. Journal of Pakistan Association of Dermatologists, 2014. **24**(4): p. 286-291. 1340-1341-1342
67. Patel, K., A. Gin, and L. Scardamaglia, *Palmoplantar keratosis caused by arsenic toxicity*. The Medical Journal of Australia, 2021. **214**(6): p. 258-258. 1343-1344
68. Richir, M.C., & Langenhorst, B. L. A. M. , *Een vrouw met 'jaarringen' op haar nagels*. Nederlands Tijdschrift voor Geneeskunde, 2011. **155**(45): p. 34-37. 1345-1346
69. Li, Y., et al., *Chronic arsenic poisoning probably caused by arsenic-based pesticides: findings from an investigation study of a household*. International journal of environmental research and public health, 2016. **13**(1): p. 133. 1347-1348-1349
70. Das, A., A. Toshniwal, and K. Majumdar, *Rain drop pigmentation in chronic arsenic poisoning*. Pigment International, 2020. **7**(1): p. 61-62. 1350-1351
71. 0013-, I.-D.-Q., *Guideline for Elemental Impurities Q3D*. ICH, 2014: p. 1-75. 1352
72. Tchounwou, P.B., et al., *Heavy metal toxicity and the environment*. Exp Suppl, 2012. **101**: p. 133-64. 1353
73. Assi, M.A., et al., *The detrimental effects of lead on human and animal health*. Vet World, 2016. **9**(6): p. 660-71. 1354-1355
74. Hernberg, S., *Lead poisoning in a historical perspective*. American journal of industrial medicine, 2000. **38**(3): p. 244-254. 1356-1357
75. Waldron, H., *James Hardy and the Devonshire colic*. Medical history, 1969. **13**(1): p. 74-81. 1358
76. FELTON, J.S., *Man, Medicine, and Work in America: A Historical Series: III. Benjamin Franklin and His Awareness of Lead Poisoning*. Journal of Occupational Medicine, 1967. **9**(11): p. 543-554. 1359-1360
77. Bellinger, D.C. and A.M. Bellinger, *Childhood lead poisoning: the torturous path from science to policy*. The Journal of clinical investigation, 2006. **116**(4): p. 853-857. 1361-1362
78. Smith, H.D., *Pediatric lead poisoning*. Archives of Environmental Health: An International Journal, 1964. **8**(2): p. 256-261. 1363-1364
79. Chisolm, J.J., Jr. and H.E. Harrison, *The exposure of children to lead*. Pediatrics, 1956. **18**(6): p. 943-58. 1365
80. Berg, J.M. and M. Zappella, *LEAD POISONING IN CHILDHOOD WITH PARTICULAR REFERENCE TO PICA AND MENTAL SEQUELAE*. J Ment Defic Res, 1964. **8**: p. 44-53. 1366-1367
81. Carter, T., *Diseases of occupations-a short history of their recognition and prevention*. Hunter's Diseases of Occupations. New York: Oxford University Press, 000, 2000. 1368-1369
82. Rubin, R., D.S. Strayer, and E. Rubin, *Rubin's pathology: clinicopathologic foundations of medicine*. 2008: Lippincott Williams & Wilkins. 1370-1371
83. Goldstein, G.W., *Neurologic concepts of lead poisoning in children*. Pediatric Annals, 1992. **21**(6): p. 384-388. 1372-1373
84. Lidsky, T.I. and J.S. Schneider, *Lead neurotoxicity in children: basic mechanisms and clinical correlates*. Brain, 2003. **126**(1): p. 5-19. 1374-1375
85. Ab Latif Wani, A.A. and J.A. Usmani, *Lead toxicity: a review*. Interdisciplinary toxicology, 2015. **8**(2): p. 55. 1376-1377
86. Lead, I.I., *Environmental Health Criteria 165. International Programme on Chemical Safety, Vol. 38*. World Health Organization, 1995: p. 93-99. 1378-1379

87. Geraldine, M. and T. Venkatesh, *Evaluation, diagnosis, and treatment of lead poisoning in a patient with occupational lead exposure: a case presentation*. Journal of Occupational medicine and Toxicology, 2007. **2**(1): p. 1-4. 1380-1382
88. Smitherman, J. and P. Harber, *A case of mistaken identity: herbal medicine as a cause of lead toxicity*. American journal of industrial medicine, 1991. **20**(6): p. 795-798. 1383-1384
89. Pearce, J., *Burton's line in lead poisoning*. European neurology, 2007. **57**(2): p. 118-119. 1385
90. Sokolowski, M.J. and G. Sisson, *Systemic lead poisoning due to an intra-articular bullet*. 2005, Slack Incorporated Thorofare, NJ. p. 411-412. 1386-1387
91. Henretiz F.M. , L.R.G., N.E. Flomenbaum and N.A. Lewis et al. editor, *Goldfrank's toxicological emergencies*, A.a. Lange, Editor. 1998. 1388-1389
92. Chawla, M. and D. Sundriyal, *Burton's Line*. The New England journal of medicine, 2012. **367**(10): p. 367-937. 1390-1391
93. Verma, S., et al., *Gastrointestinal: An oral clue to an unusual cause of pain abdomen: Burton's line*. Journal of Gastroenterology and Hepatology, 2019. **34**(4): p. 629-629. 1392-1393
94. Lau, K.K., et al., *Hong Kong poison information centre: annual report 2017*. Hong Kong Journal of Emergency Medicine, 2018. **25**(6): p. 313-323. 1394-1395
95. Chouhan, S. and S.J. Flora, *Arsenic and fluoride: two major ground water pollutants*. Indian J Exp Biol, 2010. **48**(7): p. 666-78. 1396-1397
96. Fejerskov, O., F. Manji, and V. Baelum, *The nature and mechanisms of dental fluorosis in man*. Journal of dental research, 1990. **69**(2_suppl): p. 692-700. 1398-1399
97. Robinson, C., et al., *The effect of fluoride on the developing tooth*. Caries research, 2004. **38**(3): p. 268-276. 1400
98. Aoba, T. and O. Fejerskov, *Dental fluorosis: chemistry and biology*. Critical Reviews in Oral Biology & Medicine, 2002. **13**(2): p. 155-170. 1401-1402
99. Pei, J., et al., *Fluoride decreased osteoclastic bone resorption through the inhibition of NFATc1 gene expression*. Environ Toxicol, 2014. **29**(5): p. 588-95. 1403-1404
100. Liu, Q., et al., *Analysis of the Role of Insulin Signaling in Bone Turnover Induced by Fluoride*. Biol Trace Elem Res, 2016. **171**(2): p. 380-390. 1405-1406
101. Teotia, S.P.S., et al. *Highlights of Forty Years of Research on Endemic Skeletal Fluorosis in India*. 1407
102. Levy, S.M. and N. Guha - Chowdhury, *Total fluoride intake and implications for dietary fluoride supplementation*. Journal of public health dentistry, 1999. **59**(4): p. 211-223. 1408-1409
103. Gupta, N., N. Gupta, and P. Chhabra, *Image Diagnosis: Dental and Skeletal Fluorosis*. Perm J, 2016. **20**(1): p. e105-6. 1410-1411
104. Lennon, M.A., *One in a million: the first community trial of water fluoridation*. Bulletin of the World Health Organization, 2006. **84**: p. 759-760. 1412-1413
105. Szpunar, S.M. and B. Burt, *Dental caries, fluorosis, and fluoride exposure in Michigan schoolchildren*. Journal of dental research, 1988. **67**(5): p. 802-806. 1414-1415
106. *Centers for Disease Control: Ten Great Public Health Achievements in the 20th Century*. . Washington, CDC, 2008. 1416-1417
107. McDonagh, M.S., et al., *Systematic review of water fluoridation*. Bmj, 2000. **321**(7265): p. 855-859. 1418
108. Burt, B.A., *Fluoridation and social equity*. Journal of public health dentistry, 2002. **62**(4): p. 195-200. 1419
109. Taves, D.R., *Dietary intake of fluoride ashed (total fluoride) v. unashed (inorganic fluoride) analysis of individual foods*. British Journal of Nutrition, 1983. **49**(3): p. 295-301. 1420-1421

110. Kurdi, M.S., *Chronic fluorosis: The disease and its anaesthetic implications*. Indian J Anaesth, 2016. **60**(3): p. 157-62. 1422
111. World Health Organization: *Water for Life: Making It Happen*. Geneva, WHO, 2005. 1423
112. DenBesten, P. and W. Li, *Chronic fluoride toxicity: dental fluorosis*. Fluoride and the oral environment, 2011. **22**: p. 81-96. 1424
113. Aguilar-Díaz, F.D.C., et al., *Prevalence of dental fluorosis in Mexico 2005-2015: a literature review*. Salud Publica Mex, 2017. **59**(3): p. 306-313. 1425
114. Hong, L., et al., *Timing of fluoride intake in relation to development of fluorosis on maxillary central incisors*. Community dentistry and oral epidemiology, 2006. **34**(4): p. 299-309. 1426
115. Hong, L., et al., *Fluoride intake levels in relation to fluorosis development in permanent maxillary central incisors and first molars*. Caries research, 2006. **40**(6): p. 494-500. 1427
116. A, K., *Perikymata (in German)*. Dtsch Zahnärztl Z. 1966: p. 1: 1397-1401. 1428
117. Moller, I., *Fluorides and dental fluorosis*. International dental journal, 1982. **32**(2): p. 135-147. 1429
118. Smith, G.E., *Fluoride, teeth and bone*. Medical Journal of Australia, 1985. **143**(7): p. 283-286. 1430
119. Crawford, P.J. and M.J. Aldred, *X-linked amelogenesis imperfecta: presentation of two kindreds and a review of the literature*. Oral surgery, oral medicine, oral pathology, 1992. **73**(4): p. 449-455. 1431
120. Fejerskov, O., *Dental fluorosis: a handbook for health workers*. 1988. 1432
121. Joshi, S., et al., *Skeletal fluorosis due to excessive tea and toothpaste consumption*. Osteoporos Int, 2011. **22**(9): p. 2557-60. 1433
122. Arlappa N, A.I., Srinivas R. , *Fluorosis in India: an overview*. . Int J Res Dev Health. , 2013. **1**: p. 97–102. 1434
123. Pramanik, S. and D. Saha, *The genetic influence in fluorosis*. Environ Toxicol Pharmacol, 2017. **56**: p. 157-162. 1435
124. Sellami, M., et al., *Skeletal fluorosis: don't miss the diagnosis!* Skeletal Radiol, 2020. **49**(3): p. 345-357. 1436
125. Khairuddin, M.N.I., P.M.B.u.A. Iskanderdzulkarnein, and M.H.M. Halil, *Resin Infiltration Technique as Minimal Invasive Approach Towards Mild to Moderate Dental Fluorosis in an Adolescent: a Case Report*. IIUM Journal of Orofacial and Health Sciences, 2021. **2**(2): p. 63-72. 1437
126. Gerrie, A., et al., *Fluorosis detected by trephine biopsy*. British journal of haematology, 2009. **144**(3): p. 278. 1438
127. Nayak, B., M.M. Roy, and D. Chakraborti, *Dental fluorosis*. Clin Toxicol (Phila), 2009. **47**(4): p. 355. 1439
128. Stephanie V. Sherman, M.D., Editor, *Skeletal Fluorosis*. The New England Journal of Medicine **385**:16. 1440
129. Cunningham, C.G., et al., *Quantitative mineral resource assessment of copper, molybdenum, gold, and silver in undiscovered porphyry copper deposits in the Andes Mountains of South America*. USGS Open-File report, 2008. **1253**. 1441
130. Berger, B.R., et al., *Preliminary model of porphyry copper deposits*. US geological survey open-file report, 2008. **1321**: p. 55. 1442
131. Cox, D.P., et al., *Sediment-hosted copper deposits of the world: Deposit models and database*. 2007: Citeseer. 1443
132. Doebrich, J.L., *Copper: a metal for the ages*. 2009: US Department of the Interior, US Geological Survey. 1444
133. Gamakaranage, C.S., et al., *Complications and management of acute copper sulphate poisoning; a case discussion*. J Occup Med Toxicol, 2011. **6**(1): p. 34. 1445
134. Peña, M.M., J. Lee, and D.J. Thiele, *A delicate balance: homeostatic control of copper uptake and distribution*. The Journal of nutrition, 1999. **129**(7): p. 1251-1260. 1446

135. Rotilio, G., et al., *Copper - dependent oxidative stress and neurodegeneration*. IUBMB life, 2000. **50**(4 - 5): p. 309-314. 1464 1465
136. Gu, M., et al., *Oxidative-phosphorylation defects in liver of patients with Wilson's disease*. The Lancet, 2000. **356**(9228): p. 469-474. 1466 1467
137. Fuentealba, I.C. and E.M. Aburto, *Animal models of copper-associated liver disease*. Comp Hepatol, 2003. **2**(1): p. 5. 1468 1469
138. Chowdhury, A., S. Ghosh, and D. Pal, *Acute copper sulphate poisoning*. Journal of the Indian Medical Association, 1961. **36**: p. 330-336. 1470 1471
139. Nastoulis, E., et al., *Greenish-blue gastric content: Literature review and case report on acute copper sulphate poisoning*. Forensic Sci Rev, 2017. **29**(1): p. 77-91. 1472 1473
140. Gow, P.J., et al., *Diagnosis of Wilson's disease: an experience over three decades*. Gut, 2000. **46**(3): p. 415-419. 1474 1475
141. Bull, P.C., et al., *The Wilson disease gene is a putative copper transporting P-type ATPase similar to the Menkes gene*. Nature genetics, 1993. **5**(4): p. 327-337. 1476 1477
142. Petrukhin, K., et al., *Mapping, cloning and genetic characterization of the region containing the Wilson disease gene*. Nature genetics, 1993. **5**(4): p. 338-343. 1478 1479
143. Tanzi, R., et al., *The Wilson disease gene is a copper transporting ATPase with homology to the Menkes disease gene*. Nature genetics, 1993. **5**(4): p. 344-350. 1480 1481
144. Merle, U., et al., *Truncating mutations in the Wilson disease gene ATP7B are associated with very low serum ceruloplasmin oxidase activity and an early onset of Wilson disease*. BMC gastroenterology, 2010. **10**(1): p. 1-6. 1482 1484
145. Thomas, G.R., et al., *The Wilson disease gene: spectrum of mutations and their consequences*. Nature genetics, 1995. **9**(2): p. 210-217. 1485 1486
146. Cozma, I., C. Atherley, and N. James, *Influence of ethnic origin on the incidence of keratoconus and associated atopic disease in Asian and white patients*. Eye, 2005. **19**(8): p. 924-925. 1487 1488
147. Fleischer, B., *Zwei weitere Falle von grunlicher Verfärbung der Kornea*. Klin Monatsbl Augenheilk, 1903. **41**: p. 489-491. 1489 1490
148. Kayser, B., *Über einen Fall von angeborener grünlicher Verfärbung der Cornea*. Klin Monatsbl Augenheilkd, 1902. **40**: p. 22-25. 1491 1492
149. Gerlach W, R.W., *Besteht das Pigmet des Kayser-Fleischerschen Hornhauttringes aus silber?* Klin Wochenschr 1949. **13**: p. 48-9. 1493 1494
150. Bandmann, O., K.H. Weiss, and S.G. Kaler, *Wilson's disease and other neurological copper disorders*. The Lancet Neurology, 2015. **14**(1): p. 103-113. 1495 1496
151. Dusek, P., T. Litwin, and A. Czlonkowska, *Wilson disease and other neurodegenerations with metal accumulations*. Neurologic clinics, 2015. **33**(1): p. 175-204. 1497 1498
152. Kasper, D., et al., *Harrison's principles of internal medicine, 19e*. Vol. 1. 2015: Mcgraw-hill New York, NY, USA:.. 1499 1500
153. Cairns, J.E., H.P. Williams, and J.M. Walshe, *"Sunflower Cataract" In Wilson's Disease*. The British Medical Journal, 1969. **3**(5662): p. 95-96. 1501 1502
154. Langwińska-Wośko, E., et al., *The sunflower cataract in Wilson's disease: pathognomonic sign or rare finding?* Acta Neurologica Belgica, 2016. **116**(3): p. 325-328. 1503 1504

155. Wiebers, D.O., R. Hollenhorst, and N. Goldstein. *The ophthalmologic manifestations of Wilson's disease.* in *Mayo Clinic Proceedings*. 1977. 1505
1506
156. Siemerling E, O.H., *Pseudoskleroses (Westphal Strumpell) mit Cornealring (Kayser–Fleischer) und* 1507
doppelseitiger Scheinkatarakt, die nur bei seitlicher Beleuchtung sichtbar ist und die der nach Verletzung 1508
durch Kupfersplitter entstehenden Katarakt anlich ist. *Klin Wochenschr* 1922. **1**: p. 1087–1089. 1509
157. Roberts, E.A. and M.L. Schilsky, *Diagnosis and treatment of Wilson disease: an update.* *Hepatology*, 1510
2008. **47**(6): p. 2089-2111. 1511
158. Liver, E.A.F.T.S.O.T., *EASL clinical practice guidelines: Wilson's disease.* *Journal of hepatology*, 2012. 1512
56(3): p. 671-685. 1513
159. Walshe, J., *The eye in Wilson disease.* *QJM: An International Journal of Medicine*, 2011. **104**(5): p. 451- 1514
453. 1515
160. Goyal, V. and M. Tripathi, *Sunflower cataract in Wilson's disease.* *Journal of Neurology, Neurosurgery* 1516
& Psychiatry, 2000. **69**(1): p. 133-133. 1517
161. Huo, L.-J., R.-D. Liao, and X.-M. Chen, *Ophthalmic manifestations of Wilson's disease.* [*Zhonghua yan* 1518
ke za zhi] *Chinese journal of ophthalmology*, 2008. **44**(2): p. 128-130. 1519
162. Lössner, A., et al., *The Kayser-Fleischer ring during long-term treatment in Wilson's disease* 1520
(hepatolenticular degeneration). *Graefe's archive for clinical and experimental ophthalmology*, 1986. 1521
224(2): p. 152-155. 1522
163. Hsieh, Y.-C. and Y.-Y. Tsai, *Sunflower cataract.* *Postgraduate medical journal*, 2016. **92**(1088): p. 366- 1523
366. 1524
164. Arora, N., et al., *Kayser-Fleischer ring.* *Qjm*, 2020. **113**(5): p. 361. 1525
165. Perea Pérez, R., et al., *Kayser-Fleischer rings.* *Med Clin (Barc)*, 2018. **150**(5): p. 207. 1526
166. Bigdon, E., et al., *[Diagnosis of Kayser Fleischer Ring: Can Early Diagnosis Improve the Outcome of* 1527
Wilson's Disease?]. *Klin Monbl Augenheilkd*, 2020. **237**(10): p. 1237-1239. 1528
167. Ram, J. and A. Gupta, *Kayser-Fleischer ring and sunflower cataract in Wilson disease.* *JAMA* 1529
ophthalmology, 2014. **132**(7): p. 873-873. 1530
168. Franke, K.W., *A new toxicant occurring naturally in certain samples of plant foodstuffs. 1. Results obtained* 1531
in preliminary feeding trials. *Journal of Nutrition*, 1934. **8**: p. 597-608. 1532
169. Moxon, A.L., *Alkali disease or selenium poisoning.* 1937. 1533
170. Reilly, C., *The biology of selenium.* *Selenium in Food and Health*, 2006: p. 20-42. 1534
171. Organization, W.H., *Trace elements in human nutrition and health.* 1996: World Health Organization. 1535
172. (ATSDR)., A.f.T.S.a.D.R., *Toxicological profile for Selenium.* . Atlanta, GA: U.S. Department of Health 1536
and Human Services, Public Health Service., 2003. 1537
173. (IOM)., I.o.M., *Selenium. In: Dietary Reference Intakes for Vitamin C, Vitamin E, Selenium, and* 1538
Carotenoids. Washington, DC: National Academy Press;, 2000: p. 284-324. 1539
174. Levander, O.A., *Scientific rationale for the 1989 recommended dietary allowance for selenium.* *Journal of* 1540
the American Dietetic Association, 1991. **91**(12): p. 1572-1576. 1541
175. Fordyce, F.M., *Selenium deficiency and toxicity in the environment,* in *Essentials of medical geology.* 2013, 1542
Springer. p. 375-416. 1543
176. WHO, *Selenium in drinking-water. Background document for development of WHO Guidelines for* 1544
Drinking-water Quality. (WHO/HSE/WSH/10.01/14). WHO 2011. 1545

177. Civil, I. and M. McDonald, *Acute selenium poisoning: case report*. The New Zealand Medical Journal, 1978. **87**(612): p. 354-356. 1546
1547
178. Alderman, L.C. and J.J. Bergin, *Hydrogen selenide poisoning: an illustrative case with review of the literature*. Archives of Environmental Health: An International Journal, 1986. **41**(6): p. 354-358. 1548
1549
179. Sunde, R.A., *Selenium In: Shils ME, Shike M, Ross CA, Caballero B, Cousins RJ, editors. Modern Nutrition in Health and Disease*. 2012, Philadelphia: Lippincott Williams and Wilkins. 1550
1551
180. Bopp, B.A., R.C. Bonders, and J.W. Kesterson, *Metabolic fate of selected selenium compounds in laboratory animals and man*. Drug metabolism reviews, 1982. **13**(2): p. 271-318. 1552
1553
181. Brown, D.G. and R.F. Burk, *Selenium retention in tissues and sperm of rats fed a torula yeast diet*. The Journal of nutrition, 1973. **103**(1): p. 102-108. 1554
1555
182. Thomassen Y, A.J., *Selenium in human tissues*. In: Ihnat M, editor. *Occurrence and Distribution of Selenium*. Boca Raton, FL: CRC Press; 1986. 1556
1557
183. Spallholz, J.E., *On the nature of selenium toxicity and carcinostatic activity*. Free Radical Biology and Medicine, 1994. **17**(1): p. 45-64. 1558
1559
184. Nuttall, K.L., *Evaluating selenium poisoning*. Annals of Clinical & Laboratory Science, 2006. **36**(4): p. 409-420. 1560
1561
185. Suzuki, K. and Y. Ogra, *Metabolic pathway for selenium in the body: speciation by HPLC-ICP MS with enriched Se*. Food Additives & Contaminants, 2002. **19**(10): p. 974-983. 1562
1563
186. Burk, R.F., K.E. Hill, and A.K. Motley, *Selenoprotein metabolism and function: evidence for more than one function for selenoprotein P*. The Journal of nutrition, 2003. **133**(5): p. 1517S-1520S. 1564
1565
187. Lockitch, G., *Selenium: clinical significance and analytical concepts*. Critical reviews in clinical laboratory sciences, 1989. **27**(6): p. 483-541. 1566
1567
188. Hunsaker, D.M., H.A. Spiller, and D. Williams, *Acute selenium poisoning: suicide by ingestion*. Journal of Forensic Science, 2005. **50**(4): p. JFS2004247-5. 1568
1569
189. Salonen, J., et al., *Association between cardiovascular death and myocardial infarction and serum selenium in a matched-pair longitudinal study*. The Lancet, 1982. **320**(8291): p. 175-179. 1570
1571
190. Manati, W., et al., *Protective role of selenium supplementation against cardiac lesions induced by the combination of levomepromazine and risperidone in the rabbit*. Human & experimental toxicology, 2009. **28**(8): p. 461-467. 1572
1573
1574
191. Ip, C., *The chemopreventive role of selenium in carcinogenesis*. Adv Exp Med Biol, 1986. **206**: p. 431-47. 1575
192. Clark, R.F., et al., *Selenium poisoning from a nutritional supplement*. Jama, 1996. **275**(14): p. 1087-1088. 1576
193. Dara, L., J. Hewett, and J.K. Lim, *Hydroxycut hepatotoxicity: a case series and review of liver toxicity from herbal weight loss supplements*. World journal of gastroenterology: WJG, 2008. **14**(45): p. 6999. 1577
1578
194. Haller, C.A., K.H. Meier, and K.R. Olson, *Seizures reported in association with use of dietary supplements*. Clinical toxicology, 2005. **43**(1): p. 23-30. 1579
1580
195. Schoepfer, A.M., et al., *Herbal does not mean innocuous: ten cases of severe hepatotoxicity associated with dietary supplements from Herbalife® products*. Journal of hepatology, 2007. **47**(4): p. 521-526. 1581
1582
196. Center for Food Safety and Applied Nutrition, U.F.a.D.A., *Overview of dietary supplements*. 2009: <http://www.fda.gov/Food/DietarySupplements/ConsumerInformation/ucm110417.htm>. 1583
1584
197. Schellmann, B., H. Raithel, and K. Schaller, *Acute fatal selenium poisoning*. Archives of toxicology, 1986. **59**(1): p. 61-63. 1585
1586

198. Chan, S., B. Gerson, and S. Subramaniam, *The role of copper, molybdenum, selenium, and zinc in nutrition and health*. Clinics in laboratory medicine, 1998. **18**(4): p. 673-685.
199. Pentel, P., D. Fletcher, and J. Jentzen, *Fatal acute selenium toxicity*. Journal of Forensic Science, 1985. **30**(2): p. 556-562.
200. Ruta, D. and S. Haider, *Attempted murder by selenium poisoning*. BMJ: British Medical Journal, 1989. **299**(6694): p. 316.
201. Jensen, R., *Selenium intoxication-New York*. Morbid. Mortal. Weekly Rep., 1984. **33**: p. 157-158.
202. Yang, G., et al., *Endemic selenium intoxication of humans in China*. The American journal of clinical nutrition, 1983. **37**(5): p. 872-881.
203. Stadtman, T.C., *Selenium biochemistry*. Science, 1974. **183**(4128): p. 915-22.
204. Stárka, L., et al., *Hormonal profile of men with premature balding*. Experimental and clinical endocrinology & diabetes, 2004. **112**(01): p. 24-28.
205. Joint, F., *Vitamin and mineral requirements in human nutrition*. 2004: Diamond Pocket Books (P) Ltd.
206. Agarwal, P., S. Sharma, and U. Agarwal, *Selenium toxicity: A rare diagnosis*. Indian journal of dermatology, venereology and leprology, 2016. **82**(6).
207. Senthilkumaran, S., et al., *Paradise nut paradox: alopecia due to selenosis from a nutritional therapy*. International journal of trichology, 2012. **4**(4): p. 283.
208. Gilbertson, K., et al., *Scalp discoloration from selenium sulfide shampoo: a case series and review of the literature*. Pediatric dermatology, 2012. **29**(1): p. 84-88.
209. Aldosary, B.M., et al., *Case series of selenium toxicity from a nutritional supplement*. Clinical Toxicology, 2012. **50**(1): p. 57-64.
210. Goldman, L.R., M.W. Shannon, and C.o.E. Health, *Technical report: mercury in the environment: implications for pediatricians*. Pediatrics, 2001. **108**(1): p. 197-205.
211. Amin-Zaki, L., et al., *Methylmercury poisoning in Iraqi children: clinical observations over two years*. Br Med J, 1978. **1**(6113): p. 613-616.
212. Carman, K.B., et al., *Acute mercury poisoning among children in two provinces of Turkey*. European journal of pediatrics, 2013. **172**(6): p. 821-827.
213. Gul Oz, S., et al., *Mercury vapor inhalation and poisoning of a family*. Inhalation toxicology, 2012. **24**(10): p. 652-658.
214. Maurissen, J.P., *History of mercury and mercurialism*. NY State J. Med.:(United States), 1981. **81**(13).
215. Sunderman, F.W., *Perils of mercury*. Ann Clin Lab Sci, 1988. **18**(2): p. 89-101.
216. Goldwater, L.J., *From Hippocrates to Ramazzini: early history of industrial medicine*. Annals of Medical History, 1936. **8**(1): p. 27.
217. Goldwater, L.J., *Mercury: a history of quicksilver*. Hat industry. In: Goldwater LJ, editor. Baltimore (MD): York Press, 1955.
218. O'shea, J., *'Two minutes with venus, two years with mercury'-mercury as an antisyphilitic chemotherapeutic agent*. Journal of the Royal Society of Medicine, 1990. **83**(6): p. 392-395.
219. Advisory, F.C., *An important message for pregnant women and women of childbearing age who may become pregnant about the risks of mercury in fish*. FDA Newsletter, 2001.
220. Shenker, B.J., T.L. Guo, and I.M. Shapiro, *Low-level methylmercury exposure causes human T-cells to undergo apoptosis: evidence of mitochondrial dysfunction*. Environ Res, 1998. **77**(2): p. 149-59.

221. Glezos, J.D., J.E. Albrecht, and R.D. Gair, *Pneumonitis after inhalation of mercury vapours*. Can Respir J, 2006. **13**(3): p. 150-2. 1628
222. Risher, J., *Toxicological profile for mercury*. 1999. 1629
223. Graeme, K.A. and C.V. Pollack Jr, *Heavy metal toxicity, part I: arsenic and mercury*. The Journal of emergency medicine, 1998. **16**(1): p. 45-56. 1630
224. Risher, J.F. and S.N. Amler, *Mercury exposure: evaluation and intervention: the inappropriate use of chelating agents in the diagnosis and treatment of putative mercury poisoning*. Neurotoxicology, 2005. **26**(4): p. 691-699. 1631
225. Lide, D.R., *CRC handbook of chemistry and physics, internet version 2005*. 2005, CRC Press, Boca Raton, FL. 1632
226. Boyd, A.S., et al., *Mercury exposure and cutaneous disease*. Journal of the American Academy of Dermatology, 2000. **43**(1): p. 81-90. 1633
227. Chang, L., *Mercury neurotoxicity: Effects and mechanisms*. Handbook of neurotoxicology, 1995: p. 31-59. 1634
228. Seiler, H.G., H. Sigel, and A. Sigel, *Handbook on toxicity of inorganic compounds*. 1988. 1635
229. Hursh, J.B., et al., *Clearance of mercury (Hg-197, Hg-203) vapor inhaled by human subjects*. Archives of Environmental Health: An International Journal, 1976. **31**(6): p. 302-309. 1636
230. Song, Y. and A. Li, *Massive elemental mercury ingestion*. Clinical toxicology, 2007. **45**(2): p. 193-193. 1637
231. Rusyniak, D.E. and K.A. Nañagas, *Conservative management of elemental mercury retained in the appendix*. Clinical Toxicology, 2008. **46**(9): p. 831-833. 1638
232. Gutiérrez, F. and L. Leon, *Elemental mercury embolism to the lung*. New England Journal of Medicine, 2000. **342**(24): p. 1791-1791. 1639
233. Rowens, B., et al., *Respiratory failure and death following acute inhalation of mercury vapor: a clinical and histologic perspective*. Chest, 1991. **99**(1): p. 185-190. 1640
234. Aguado, S., et al., *Acute mercury vapour intoxication: report of six cases*. Nephrology Dialysis Transplantation, 1989. **4**(2): p. 133-136. 1641
235. Lilis, R., A. Miller, and Y. Lerman, *Acute mercury poisoning with severe chronic pulmonary manifestations*. Chest, 1985. **88**(2): p. 306-9. 1642
236. Clarkson, T.W., L. Magos, and G.J. Myers, *The toxicology of mercury—current exposures and clinical manifestations*. New England Journal of Medicine, 2003. **349**(18): p. 1731-1737. 1643
237. Tunnessen, W.W., K.J. McMahon, and M. Baser, *Acrodynia: exposure to mercury from fluorescent light bulbs*. Pediatrics, 1987. **79**(5): p. 786-789. 1644
238. Warkany, J. and D.M. Hubbard, *Adverse mercurial reactions in the form of acrodynia and related conditions*. AMA American journal of diseases of children, 1951. **81**(3): p. 335-373. 1645
239. Curtis, H., et al., *Mercury as a health hazard*. Archives of disease in childhood, 1987. **62**(3): p. 293-295. 1646
240. Mercer, J.J., L. Bercovitch, and J.J. Muglia, *Acrodynia and hypertension in a young girl secondary to elemental mercury toxicity acquired in the home*. Pediatr Dermatol, 2012. **29**(2): p. 199-201. 1647
241. Nakayama H, S.M., Hada S. , *Mercury exanthem*. . J Am Acad Dermatol., 1984. **11**: p. 137-139. 1648
242. Sedano, H.O., *Mercury poisoning and acrodynia*. Oral surgery, oral medicine, oral pathology, oral radiology, and endodontics, 1998. **85**(4): p. 349-349. 1649
243. Henningson, C., et al., *Acute mercury poisoning (acrodynia) mimicking pheochromocytoma in an adolescent*. The Journal of pediatrics, 1993. **122**(2): p. 252-253. 1650

244. Torres, A.D., A.N. Rai, and M.L. Hardiek, *Mercury intoxication and arterial hypertension: report of two patients and review of the literature*. Pediatrics, 2000. **105**(3): p. e34-e34. 1670-1671
245. Wossmann, W., et al., *Mercury intoxication presenting with hypertension and tachycardia*. Archives of disease in childhood, 1999. **80**(6): p. 556-557. 1672-1673
246. Beuter, A. and A. De Geoffroy, *Can tremor be used to measure the effect of chronic mercury exposure in human subjects?* Neurotoxicology, 1996. **17**(1): p. 213-227. 1674-1675
247. Netterstrøm, B., B. Guldager, and J. Heebøll, *Acute mercury intoxication examined with coordination ability and tremor*. Neurotoxicology and Teratology, 1996. **18**(4): p. 505-509. 1676-1677
248. Buckell, M., D. Hunter, and et al., *Chronic mercury poisoning*. British journal of industrial medicine, 1946. **3**(2): p. 55-63. 1678-1679
249. Bernhoft, R.A., *Mercury toxicity and treatment: a review of the literature*. Journal of environmental and public health, 2012. **2012**. 1680-1681
250. Smith, R., et al., *Effects of exposure to mercury in the manufacture of chlorine*. American Industrial Hygiene Association Journal, 1970. **31**(6): p. 687-700. 1682-1683
251. Moneim, R.A.A., M. El Deeb, and A.A. Rabea, *Gingival pigmentation (cause, treatment and histological preview)*. Future Dental Journal, 2017. **3**(1): p. 1-7. 1684-1685
252. Park, J.-D. and W. Zheng, *Human exposure and health effects of inorganic and elemental mercury*. Journal of preventive medicine and public health, 2012. **45**(6): p. 344. 1686-1687
253. Broussard, L.A., et al., *The toxicology of mercury*. Laboratory medicine, 2002. **33**(8): p. 614-625. 1688
254. De Bont, B., et al., *Yellow mercuric oxide ointment and mercury intoxication*. European journal of pediatrics, 1986. **145**(3): p. 217-218. 1689-1690
255. Dargan, P.I., et al., *Case report: severe mercuric sulphate poisoning treated with 2, 3-dimercaptopropane-1-sulphonate and haemodiafiltration*. Critical Care, 2003. **7**(3): p. 1-6. 1691-1692
256. Harada, M., *Minamata disease: methylmercury poisoning in Japan caused by environmental pollution*. Critical reviews in toxicology, 1995. **25**(1): p. 1-24. 1693-1694
257. Myers, G.J. and P.W. Davidson, *Prenatal methylmercury exposure and children: neurologic, developmental, and behavioral research*. Environmental Health Perspectives, 1998. **106**(suppl 3): p. 841-847. 1695-1696-1697
258. Aberg, B., et al., *Metabolism of methyl mercury (^{203}Hg) compounds in man: Excretion and distribution*. Archives of Environmental Health: An International Journal, 1969. **19**(4): p. 478-484. 1698-1699
259. Berlin, M., J. Carlson, and T. Norseth, *Dose-dependence of methylmercury metabolism: A study of distribution: Biotransformation and excretion in the squirrel monkey*. Archives of Environmental Health: An International Journal, 1975. **30**(6): p. 307-313. 1700-1702
260. Baker, J.P., *Mercury, vaccines, and autism: one controversy, three histories*. American journal of public health, 2008. **98**(2): p. 244-253. 1703-1704
261. McNeil, N., et al., *Domestic metallic mercury poisoning*. The Lancet, 1984. **323**(8371): p. 269-271. 1705
262. Bakir, F., et al., *Methylmercury poisoning in Iraq*. Science, 1973. **181**(4096): p. 230-241. 1706
263. Eto, K., *Minamata Disease*. Neuropathology, 20, S14-S19. 2000. 1707
264. Eto, K., et al., *An autopsy case of Minamata disease (methylmercury poisoning)—pathological viewpoints of peripheral nerves*. Toxicologic Pathology, 2002. **30**(6): p. 714-722. 1708-1709
265. Atti, S.K., et al., *All that glitters is not gold: Mercury poisoning in a family mimicking an infectious illness*. Curr Probl Pediatr Adolesc Health Care, 2020. **50**(2): p. 100758. 1710-1711

266. Do, S.Y., et al., *Cases of acute mercury poisoning by mercury vapor exposure during the demolition of a fluorescent lamp factory*. *Ann Occup Environ Med*, 2017. **29**: p. 19. 1712
1713
267. Abhilash, K.P., J.J. Arul, and D. Bala, *Fatal overdose of iron tablets in adults*. *Indian J Crit Care Med*, 2013. **17**(5): p. 311-3. 1714
1715
268. MACKENZIE, E.L., K. IWASAKI, and Y. TSUJI, *Comprehensive Invited Review*. *ANTIOXIDANTS & REDOX SIGNALING*, 2008. **10**(6). 1716
1717
269. Gerlach, M., et al., *Altered brain metabolism of iron as a cause of neurodegenerative diseases?* *J Neurochem*, 1994. **63**(3): p. 793-807. 1718
1719
270. Baranwal, A.K. and S.C. Singhi, *Acute iron poisoning: management guidelines*. *Indian pediatrics*, 2003. **40**(6): p. 534-540. 1720
1721
271. Singhi, S.C., A.K. Baranwal, and J. M, *Acute iron poisoning: clinical picture, intensive care needs and outcome*. *Indian Pediatr*, 2003. **40**(12): p. 1177-82. 1722
1723
272. Pestaner, J.P., et al., *Ferrous sulfate toxicity: a review of autopsy findings*. *Biol Trace Elem Res*, 1999. **69**(3): p. 191-8. 1724
1725
273. Madiwale, T. and E. Liebelt, *Iron: not a benign therapeutic drug*. *Curr Opin Pediatr*, 2006. **18**(2): p. 174-9. 1726
1727
274. Sane, M.R., et al., *Fatal Iron Toxicity in an Adult: Clinical Profile and Review*. *Indian J Crit Care Med*, 2018. **22**(11): p. 801-803. 1728
1729
275. Chen, M.R., et al., *Acute iron intoxication: a case report with ferric chloride ingestion*. *Zhonghua Yi Xue Za Zhi (Taipei)*, 1993. **52**(4): p. 269-72. 1730
1731
276. Lacoste, H., et al., *Acute iron intoxication in pregnancy: case report and review of the literature*. *Obstet Gynecol*, 1992. **80**(3 Pt 2): p. 500-1. 1732
1733
277. Tran, T., et al., *Intentional iron overdose in pregnancy--management and outcome*. *J Emerg Med*, 2000. **18**(2): p. 225-8. 1734
1735
278. Tran, T., et al., *Acute intentional iron overdose in pregnancy*. *Obstet Gynecol*, 1998. **92**(4 Pt 2): p. 678-80. 1736
279. Yuen, H.W.B., W., *Iron Toxicity*, in *StatPearls*. 2022, StatPearls Publishing Copyright © 2022, StatPearls Publishing LLC.: Treasure Island (FL). 1737
1738
280. Wang, W.C., N. Ahmed, and M. Hanna, *Non-transferrin-bound iron in long-term transfusion in children with congenital anemias*. *J Pediatr*, 1986. **108**(4): p. 552-7. 1739
1740
281. Skoczynska, A., et al., *Acute iron poisoning in adult female*. *Human & experimental toxicology*, 2007. **26**(8): p. 663-666. 1741
1742
282. Lacouture, P.G., et al., *Emergency assessment of severity in iron overdose by clinical and laboratory methods*. *J Pediatr*, 1981. **99**(1): p. 89-91. 1743
1744
283. Covey, T.J., *Ferrous sulfate poisoning*. *J. Pediatr.*, 1954: p. 64:218-226. 1745
284. Pillay, V.V., *Modern Medical Toxicology*. 4th ed. 2013, New Delhi: Jaypee Brothers Medical Publishers. 1746
1747
285. Murray, M.J., *Iron absorption*. *Clin Toxicol*, 1971. **4**(4): p. 545-58. 1748
286. Liebelt EL, K.R., *Acute iron poisoning*. Available from: www.uptodate.com. 2013 Aug 07 1749
287. Simonse, E., et al., *Iron autointoxication in a 16-year-old girl: a protective role for hepcidin?* *Ann Clin Biochem*, 2013. **50**(Pt 1): p. 76-9. 1750
1751
288. Mahesh, K. and R. Rani, *A case of iron poisoning-case report*. *Int J Basic Appl Med Sci*, 2014. **4**: p. 101-3. 1752

289. Robertson, A. and M. Tenenbein, *Hepatotoxicity in acute iron poisoning*. Hum Exp Toxicol, 2005. **24**(11): p. 559-62. 1753
290. Gandhi, R.K. and F.H. Robarts, *Hour-glass stricture of the stomach and pyloric stenosis due to ferrous sulphate poisoning*. Br J Surg, 1962. **49**: p. 613-7. 1754
291. Feder, J.N., et al., *A novel MHC class I-like gene is mutated in patients with hereditary haemochromatosis*. Nat Genet, 1996. **13**(4): p. 399-408. 1755
292. Powell, L.W., R.C. Seckington, and Y. Deugnier, *Haemochromatosis*. Lancet, 2016. **388**(10045): p. 706-16. 1756
293. Bacon, B.R., et al., *Molecular medicine and hemochromatosis: at the crossroads*. Gastroenterology, 1999. **116**(1): p. 193-207. 1757
294. Brissot, P. and F. de Bels, *Current approaches to the management of hemochromatosis*. Hematology Am Soc Hematol Educ Program, 2006: p. 36-41. 1758
295. Pietrangelo, A., *Hereditary hemochromatosis--a new look at an old disease*. N Engl J Med, 2004. **350**(23): p. 2383-97. 1759
296. Drobnik J, S.R., *Hemochromatosis*. Emedicine July 21, 2005. 1760
297. Sfeir HE, K.D., *Hemochromatosis*. Emedicine June 8, 2005. 1761
298. Borgaonkar, M.R., *Hemochromatosis. More common than you think*. Canadian family physician, 2003. **49**(1): p. 36-43. 1762
299. Tavill, A.S., *Diagnosis and management of hemochromatosis*. Hepatology, 2001. **33**(5): p. 1321-8. 1763
300. Limdi, J.K. and J.R. Crampton, *Hereditary haemochromatosis*. Qjm, 2004. **97**(6): p. 315-24. 1764
301. Pietrangelo, A., *Juvenile hemochromatosis*. J Hepatol, 2006. **45**(6): p. 892-4. 1765
302. Ekanayake, D., C. Roddick, and L.W. Powell, *Recent advances in hemochromatosis: a 2015 update : a summary of proceedings of the 2014 conference held under the auspices of Hemochromatosis Australia*. Hepatol Int, 2015. **9**(2): p. 174-82. 1766
303. Brissot, P., et al., *Current approach to hemochromatosis*. Blood Rev, 2008. **22**(4): p. 195-210. 1767
304. Ajioka, R.S. and J.P. Kushner, *Clinical consequences of iron overload in hemochromatosis homozygotes*. Blood, 2003. **101**(9): p. 3351-3; discussion 3354-8. 1768
305. Allen, K.J., et al., *Iron-overload-related disease in HFE hereditary hemochromatosis*. N Engl J Med, 2008. **358**(3): p. 221-30. 1769
306. Beutler, E., et al., *Penetrance of 845G--> A (C282Y) HFE hereditary haemochromatosis mutation in the USA*. Lancet, 2002. **359**(9302): p. 211-8. 1770
307. Bulaj, Z.J., et al., *Disease-related conditions in relatives of patients with hemochromatosis*. N Engl J Med, 2000. **343**(21): p. 1529-35. 1771
308. Gurrin, L.C., et al., *The natural history of serum iron indices for HFE C282Y homozygosity associated with hereditary hemochromatosis*. Gastroenterology, 2008. **135**(6): p. 1945-52. 1772
309. Wojcik, J.P., et al., *Natural history of C282Y homozygotes for hemochromatosis*. Can J Gastroenterol, 2002. **16**(5): p. 297-302. 1773
310. Witte, D.L., et al., *Practice guideline development task force of the College of American Pathologists. Hereditary hemochromatosis*. Clin Chim Acta, 1996. **245**(2): p. 139-200. 1774
311. Pittelkow, M.R. and S. Flores, *Hemochromatosis*. <https://www.dermatologyadvisor.com/home/decision-support-in-medicine/dermatology/hemochromatosis-3/>. Accessed on 16/03/2022. 2017. 1775

312. Chmieliauskas, S., et al., *Autopsy relevance determining hemochromatosis: case report*. *Medicine*, 2017. **96**(49). 1794-1795
313. Nogawa, K. *Kanazawa Medical University Uchinada, Ishikawa, Japan*. in *Changing Metal Cycles and Human Health: Report of the Dahlem Workshop on Changing Metal Cycles and Human Health*, Berlin 1983, March 20-25. 1984. Springer. 1796-1798
314. Probst, G., *Cadmium: Absorption and excretion in mammals*. In Mennear JH (Ed): "*Cadmium Toxicity*". New York: Marcel Dekker, Inc, 1979: p. 29-59. 1799-1800
315. Świergosz, R., et al., *Accumulation of cadmium in and its effect on bank vole tissues after chronic exposure*. *Ecotoxicology and Environmental Safety*, 1998. **41**(2): p. 130-136. 1801-1802
316. Järup, L., *Hazards of heavy metal contamination*. *British medical bulletin*, 2003. **68**(1): p. 167-182. 1803
317. Godt, J., et al., *The toxicity of cadmium and resulting hazards for human health*. *Journal of occupational medicine and toxicology*, 2006. **1**(1): p. 1-6. 1804-1805
318. Johri, N., G. Jacquillet, and R. Unwin, *Heavy metal poisoning: the effects of cadmium on the kidney*. *Biometals*, 2010. **23**(5): p. 783-792. 1806-1807
319. Järup, L., et al., *Health effects of cadmium exposure—a review of the literature and a risk estimate*. *Scandinavian journal of work, environment & health*, 1998: p. 1-51. 1808-1809
320. Elinder, C.G., et al., *Cadmium in kidney cortex, liver, and pancreas from Swedish autopsies. Estimation of biological half time in kidney cortex, considering calorie intake and smoking habits*. *Arch Environ Health*, 1976. **31**(6): p. 292-302. 1810-1812
321. WHO, *Cadmium. Environmental Health Criteria*. 1992, World Health Organization: Geneva. 1813
322. Zalups, R.K. and S. Ahmad, *Molecular handling of cadmium in transporting epithelia*. *Toxicol Appl Pharmacol*, 2003. **186**(3): p. 163-88. 1814-1815
323. Wang, Y., et al., *Cadmium inhibits the electron transfer chain and induces reactive oxygen species*. *Free Radic Biol Med*, 2004. **36**(11): p. 1434-43. 1816-1817
324. Stohs, S.J., et al., *Oxidative mechanisms in the toxicity of chromium and cadmium ions*. *J Environ Pathol Toxicol Oncol*, 2000. **19**(3): p. 201-13. 1818-1819
325. Desagher, S. and J.C. Martinou, *Mitochondria as the central control point of apoptosis*. *Trends Cell Biol*, 2000. **10**(9): p. 369-77. 1820-1821
326. Thévenod, F., et al., *Up-regulation of multidrug resistance P-glycoprotein via nuclear factor-kappaB activation protects kidney proximal tubule cells from cadmium- and reactive oxygen species-induced apoptosis*. *J Biol Chem*, 2000. **275**(3): p. 1887-96. 1822-1824
327. Tzirogiannis, K.N., et al., *Time-course of cadmium-induced acute hepatotoxicity in the rat liver: the role of apoptosis*. *Arch Toxicol*, 2003. **77**(12): p. 694-701. 1825-1826
328. Svartengren, M., et al., *Distribution and concentration of cadmium in human kidney*. *Environ Res*, 1986. **39**(1): p. 1-7. 1827-1828
329. Orłowski, C. and J.K. Piotrowski, *Biological levels of cadmium and zinc in the small intestine of non-occupationally exposed human subjects*. *Hum Exp Toxicol*, 2003. **22**(2): p. 57-63. 1829-1830
330. Barbier, O., et al., *Effect of heavy metals on, and handling by, the kidney*. *Nephron Physiol*, 2005. **99**(4): p. p105-10. 1831-1832
331. Bernard, A., *Renal dysfunction induced by cadmium: biomarkers of critical effects*. *Biometals*, 2004. **17**(5): p. 519-23. 1833-1834

332. Wolff, N.A., et al., *Megalyn-dependent internalization of cadmium-metallothionein and cytotoxicity in cultured renal proximal tubule cells*. J Pharmacol Exp Ther, 2006. **318**(2): p. 782-91. 1835
333. Jin, T., et al., *Cadmium biomonitoring and renal dysfunction among a population environmentally exposed to cadmium from smelting in China (ChinaCad)*. Biometals, 2002. **15**(4): p. 397-410. 1836
334. Flanagan, P.R., et al., *Increased dietary cadmium absorption in mice and human subjects with iron deficiency*. Gastroenterology, 1978. **74**(5 Pt 1): p. 841-6. 1837
335. Horiguchi, H., et al., *Comprehensive study of the effects of age, iron deficiency, diabetes mellitus, and cadmium burden on dietary cadmium absorption in cadmium-exposed female Japanese farmers*. Toxicol Appl Pharmacol, 2004. **196**(1): p. 114-23. 1838
336. Gunshin, H., et al., *Cloning and characterization of a mammalian proton-coupled metal-ion transporter*. Nature, 1997. **388**(6641): p. 482-488. 1839
337. Seidal, K., et al., *Fatal cadmium-induced pneumonitis*. Scand J Work Environ Health, 1993. **19**(6): p. 429-31. 1840
338. Yates, D. and K. Goldman, *Acute cadmium poisoning in a foreman plater welder*. British journal of industrial medicine, 1990. **47**(6): p. 429. 1841
339. Nordberg, G.F., *Historical perspectives on cadmium toxicology*. Toxicol Appl Pharmacol, 2009. **238**(3): p. 192-200. 1842
340. Nordberg, G.F., *Cadmium and health in the 21st century--historical remarks and trends for the future*. Biometals, 2004. **17**(5): p. 485-9. 1843
341. Inaba, T., et al., *Estimation of cumulative cadmium intake causing Itai-itai disease*. Toxicol Lett, 2005. **159**(2): p. 192-201. 1844
342. Nogawa, K., et al., *[Itai-itai disease]*. Nihon Eiseigaku Zasshi, 1975. **30**(1): p. 76. 1845
343. Nogawa, K., et al., *Environmental cadmium exposure, adverse effects and preventive measures in Japan*. Biometals, 2004. **17**(5): p. 581-7. 1846
344. Kazantzis, G., *Renal tubular dysfunction and abnormalities of calcium metabolism in cadmium workers*. Environ Health Perspect, 1979. **28**: p. 155-9. 1847
345. Alfvén, T., et al., *Low-level cadmium exposure and osteoporosis*. J Bone Miner Res, 2000. **15**(8): p. 1579-86. 1848
346. Nordberg, G., et al., *Low bone density and renal dysfunction following environmental cadmium exposure in China*. Ambio, 2002. **31**(6): p. 478-81. 1849
347. Staessen, J.A., et al., *Environmental exposure to cadmium, forearm bone density, and risk of fractures: prospective population study*. Public Health and Environmental Exposure to Cadmium (PheeCad) Study Group. Lancet, 1999. **353**(9159): p. 1140-4. 1850
348. Parizek, J. and Z. Zahor, *Effect of cadmium salts on testicular tissue*. Nature, 1956. **177**(4518): p. 1036. 1851
349. Everett, C.J. and I.L. Frithsen, *Association of urinary cadmium and myocardial infarction*. Environ Res, 2008. **106**(2): p. 284-6. 1852
350. Spieker, C., W. Zidek, and H. Zunkley, *Cadmium and hypertension*. Nephron, 1987. **47 Suppl 1**: p. 34-6. 1853
351. Schwartz, G.G., D. Il'yasova, and A. Ivanova, *Urinary cadmium, impaired fasting glucose, and diabetes in the NHANES III*. Diabetes Care, 2003. **26**(2): p. 468-70. 1854
352. Wagstaff, A.J., P. Benfield, and J.P. Monk, *Colloidal bismuth subcitrate*. Drugs, 1988. **36**(2): p. 132-157. 1855
353. Gorbach, S.L., *Bismuth therapy in gastrointestinal diseases*. Gastroenterology, 1990. **99**(3): p. 863-875. 1856

354. Mohan, R., *Green bismuth*. Nature chemistry, 2010. **2**(4): p. 336-336. 1877
355. Lambert, J. and P. Midolo, *The actions of bismuth in the treatment of Helicobacter pylori infection*. Alimentary pharmacology & therapeutics, 1997. **11**(S1): p. 27-33. 1878
1879
356. Bierer, D.W., *Bismuth subsalicylate: history, chemistry, and safety*. Reviews of infectious diseases, 1990. **12**(Supplement_1): p. S3-S8. 1880
1881
357. Menge, H., et al., *Pharmacology of bismuth*. EUROPEAN JOURNAL OF GASTROENTEROLOGY & HEPATOLOGY, 1992. **4**: p. S41-S47. 1882
1883
358. Morison, R., *The treatment of infected suppurating war wounds*. Vol. 188. 1916: Lancet 1884
359. Sun, H., *Biological Chemistry of Arsenic, Antimony and Bismuth*; John Wiley & Sons: Hoboken, NJ, USA, 2011. 1885
1886
360. Marshall, B. and J.R. Warren, *Unidentified curved bacilli in the stomach of patients with gastritis and peptic ulceration*. The lancet, 1984. **323**(8390): p. 1311-1315. 1887
1888
361. Gaynor, D. and D.M. Griffith, *The prevalence of metal-based drugs as therapeutic or diagnostic agents: beyond platinum*. Dalton Transactions, 2012. **41**(43): p. 13239-13257. 1889
1890
362. Jones, J., *Bipp: a case of toxicity?* Oral surgery, oral medicine, oral pathology, 1990. **69**(6): p. 668-671. 1891
363. Macdonald, K., A. Farboud, and A. Jardine, *Bismuth iodoform paraffin paste: a review*. The Journal of Laryngology & Otology, 2012. **126**(11): p. 1192-1192. 1892
1893
364. Tan, R., et al., *Myoclonus From Intoxication by Bismuth Iodoform Paraffin Paste (BIPP) Nasopharyngeal Packing*. Cureus, 2021. **13**(10). 1894
1895
365. Vouk, V., G. Nordberg, and L. Friberg, *Handbook on the Toxicology of Metals*. 1979: Elsevier/North-Holland Biomedical Press. 1896
1897
366. Cengiz, N., et al., *Acute renal failure after overdose of colloidal bismuth subcitrate*. Pediatr Nephrol, 2005. **20**(9): p. 1355-8. 1898
1899
367. Slikkerveer, A., et al., *Comparison of enhanced elimination of bismuth in humans after treatment with meso-2,3-dimercaptosuccinic acid and D,L-2,3-dimercaptopropane-1-sulfonic acid*. Analyst, 1998. **123**(1): p. 91-2. 1900
1902
368. Phillips, R.H., et al., *Solubility, absorption, and anti-Helicobacter pylori activity of bismuth subnitrate and colloidal bismuth subcitrate: In vitro data do not predict in vivo efficacy*. Helicobacter, 2000. **5**(3): p. 176-182. 1903
1904
1905
369. Slikkerveer, A. and F.A. de Wolff, *Pharmacokinetics and toxicity of bismuth compounds*. Med Toxicol Adverse Drug Exp, 1989. **4**(5): p. 303-23. 1906
1907
370. Nwokolo, C.U., et al., *The absorption of bismuth from oral doses of tripotassium dicitrato bismuthate*. Aliment Pharmacol Ther, 1989. **3**(1): p. 29-39. 1908
1909
371. Le Quesne, P.M., *Toxic substances and the nervous system: the role of clinical observation*. J Neurol Neurosurg Psychiatry, 1981. **44**(1): p. 1-8. 1910
1911
372. Benet, L.Z., *Safety and pharmacokinetics: colloidal bismuth subcitrate*. Scand J Gastroenterol Suppl, 1991. **185**: p. 29-35. 1912
1913
373. Bridgeman, A.M. and A.C. Smith, *Iatrogenic bismuth poisoning. Case report*. Aust Dent J, 1994. **39**(5): p. 279-81. 1914
1915
374. Karelitz, S. and A.D. Freedman, *Hepatitis and nephrosis due to soluble bismuth*. Pediatrics, 1951. **8**(6): p. 772-7. 1916
1917

375. Walker, E.M., Jr., M.A. Fazekas-May, and W.R. Bowen, *Nephrotoxic and ototoxic agents*. Clin Lab Med, 1990. **10**(2): p. 323-54. 1918
1919
376. Weller, M.P., *Neuropsychiatric symptoms following bismuth intoxication*. Postgrad Med J, 1988. **64**(750): p. 308-10. 1920
1921
377. Robertson, J.F., *Mental illness or metal illness? Bismuth subgallate*. Medical Journal of Australia, 1974. **1**(22): p. 887-888. 1922
1923
378. Teepker, M., et al., *Myoclonic encephalopathy caused by chronic bismuth abuse*. Epileptic Disord, 2002. **4**(4): p. 229-33. 1924
1925
379. Erden, A., et al., *A case of bismuth intoxication with irreversible renal damage*. Int J Nephrol Renovasc Dis, 2013. **6**: p. 241-3. 1926
1927
380. Loghman-Adham, M., *Aminoaciduria and glycosuria following severe childhood lead poisoning*. Pediatr Nephrol, 1998. **12**(3): p. 218-21. 1928
1929
381. Kaizu, K. and K. Uriu, *[Tubulointerstitial injuries in heavy metal intoxications]*. Nihon Rinsho, 1995. **53**(8): p. 2052-6. 1930
1931
382. Saini, V., et al., *Pearls & Oy-sters: Bismuth neurotoxicity from use of topical bismuth dressing for burns*. Neurology, 2019. **92**(14): p. 680-681. 1932
1933
383. Burns, R., D.W. Thomas, and V.J. Barron, *Reversible encephalopathy possibly associated with bismuth subgallate ingestion*. Br Med J, 1974. **1**(5901): p. 220-3. 1934
1935
384. Youngman, L. and S. Harris, *BIPP madness; an iatrogenic cause of acute confusion*. Age Ageing, 2004. **33**(4): p. 406-7. 1936
1937
385. Sharma, R.R., et al., *Extradural application of bismuth iodoform paraffin paste causing relapsing bismuth encephalopathy: a case report with CT and MRI studies*. J Neurol Neurosurg Psychiatry, 1994. **57**(8): p. 990-3. 1938
1939
1940
386. Ovaska, H., et al., *Severe iatrogenic bismuth poisoning with bismuth iodoform paraffin paste treated with DMPS chelation*. Clin Toxicol (Phila), 2008. **46**(9): p. 855-7. 1941
1942
387. McCarthy, F.D., S. , *Oral manifestations of bismuth*. New England Journal of Medicine, 1925. **213**: p. 345-353. 1943
1944
388. Grund, J., *Erythema of the ninth day following bismuth therapy for syphilis*. Archives of Dermatology and Syphilology, 1940. **41**(6): p. 1076-1077. 1945
1946
389. Steinfield, W.M., H. , *Untersuchungen uber die toxischen und therapeutischen wirkungen des wismuths*. . Archiv fur Experimentelle Pathologie und Pharmazie, 1886. **xx**: p. 40-84. 1947
1948
390. Lohe, H.R., H., *Ein einfaches symptom zur Erkennung der wismut intoxication bei der syphilisbehandlung*. Medizinische Klinische, 1927. **23**: p. 1295. 1949
1950
391. Peters, E., *Bismuth stomatitis and albuminuria*. . American Journal of Syphilis, Gonorrhoea and Venereal Diseases, 1942. **26**: p. 84-95. 1951
1952
392. Pohanish, R.P., *Sitting's Handbook of toxic and hazardous chemicals and carcinogens*. Washington, DC, Chemical Emergency Preparedness Program, 1987. 1953
1954
393. Neville BW, D.D., Allen CM, Chi AC., *Physical and chemical injuries*. In: *Oral and maxillofacial pathology*. 4th ed. St. Louis: Elsevier Health Sciences; 2015: p. 288-9. 1955
1956
394. Borbinha, C., et al., *Bismuth encephalopathy- a rare complication of long-standing use of bismuth subsalicylate*. BMC Neurol, 2019. **19**(1): p. 212. 1957
1958

-
395. Bolan, S., et al., *Sources, distribution, bioavailability, toxicity, and risk assessment of heavy metal (loid) s in complementary medicines*. Environment international, 2017. **108**: p. 103-118. 1959
1960
396. Bachanek, T., et al., *Heavy metal poisoning in glass worker characterised by severe*. Annals of Agricultural and Environmental Medicine, 2000. **7**(1): p. 51. 1961
1962
397. Sienczuk, W., *Toksykologia*. PZWL, Warszawa, 1997. 1963
398. Rajkumar, V. and V. Gupta, *Heavy Metal Toxicity*. 2021: StatPearls Publishing, Treasure Island (FL). 1964
399. Hepler, B., C. Sutheimer, and I. Sunshine, *Role of the toxicology laboratory in suspected ingestions*. Pediatr Clin North Am, 1986. **33**(2): p. 245-60. 1965
1966
400. Kim, J.J., Y.S. Kim, and V. Kumar, *Heavy metal toxicity: An update of chelating therapeutic strategies*. J Trace Elem Med Biol, 2019. **54**: p. 226-231. 1967
1968
401. Staudinger, K.C. and V.S. Roth, *Occupational lead poisoning*. Am Fam Physician, 1998. **57**(4): p. 719-26, 731-2. 1969
1970
1971

ANEXOS

1. Reporting Guidelines (SANRA)

Scale for the Assessment of Narrative Review Articles – SANRA

Please rate the quality of the narrative review article in question, using categories 0–2 on the following scale. For each aspect of quality, please choose the option which best fits your evaluation, using categories 0 and 2 freely to imply general low and high quality. These are not intended to imply the worst or best imaginable quality.

1) Justification of the article's importance for the readership

- The importance is not justified. _____ 0
The importance is alluded to, but not explicitly justified. _____ 1
The importance is explicitly justified. _____ 2

2

2) Statement of concrete aims or formulation of questions

- No aims or questions are formulated. _____ 0
Aims are formulated generally but not concretely or in terms of clear questions. _____ 1
One or more concrete aims or questions are formulated. _____ 2

2

3) Description of the literature search

- The search strategy is not presented. _____ 0
The literature search is described briefly. _____ 1
The literature search is described in detail, including search terms and inclusion criteria. _____ 2

2

4) Referencing

- Key statements are not supported by references. _____ 0
The referencing of key statements is inconsistent. _____ 1
Key statements are supported by references. _____ 2

2

5) Scientific reasoning

(e.g., incorporation of appropriate evidence, such as RCTs in clinical medicine)

- The article's point is not based on appropriate arguments. _____ 0
Appropriate evidence is introduced selectively. _____ 1
Appropriate evidence is generally present. _____ 2

2

6) Appropriate presentation of data

(e.g., absolute vs relative risk; effect sizes without confidence intervals)

- Data are presented inadequately. _____ 0
Data are often not presented in the most appropriate way. _____ 1
Relevant outcome data are generally presented appropriately. _____ 2

2

Sumscore

12

SANRA – explanations and instructions

This scale is intended to help editors assess the quality of a narrative review article based on formal criteria accessible to the reader. It cannot cover other elements of editorial decision making such as degree of originality, topicality, conflicts of interest or the plausibility, correctness or completeness of the content itself. SANRA is an instrument for editors, authors, and reviewers evaluating individual manuscripts. It may also help editors to document average manuscript quality within their journal and researchers to document the manuscript quality, for example in peer review research. Using only three scoring options, 0, 1 and 2, SANRA is intended to provide a swift and pragmatic sum score for quality, for everyday use with real manuscripts, in a field where established quality standards have previously been lacking. It is not designed as an exact measurement of the quality of all theoretically possible manuscripts. For this reason, the extreme values (0 and 2) should be used relatively freely and not reserved only for perfect or hopeless articles.

We recommend that users test-rate a few manuscripts to familiarize themselves with the scale, before using it on the intended group of manuscripts. Ratings should assess the totality of a manuscript, including the abstract. The following comments clarify how each question is designed to be used.

Item 1 – Justification of the article's importance for the readership

Justification of importance for the readership must be seen in the context of each journal's readership.

Consider how well the manuscript outlines the clinical problem and highlights unanswered questions or evidence gaps – thoroughly (2), superficially (1), or not at all (0).

Item 2 – Statement of concrete/specific aims or formulation of questions

A good paper will propose one or more specific aims or questions which will be dealt with or topics which will be reviewed.

Please rate whether this has been done thoroughly and clearly (2), vaguely or unclearly (1), or not at all (0).

Item 3 – Description of the literature search

A convincing narrative review will be transparent about the sources of information on which the text is based. Please rate the degree to which you think this has been achieved. To achieve a rating of 2, it is not necessary to describe the literature search in as much detail as for a systematic review (searching multiple databases, including exact descriptions of search history, flowcharts, etc.), but it is necessary to specify search terms, and the types of literature included. A manuscript which only refers briefly to its literature search would score 1, while one not mentioning its methods would score 0.

Item 4 – Referencing

No manuscript references all statements. However, those that are essential for the arguments of the manuscript – “key statements” – should be backed by references in all or almost all cases. Exceptions could reasonably be made for rating purposes where a key statement has uncontroversial face-validity, such as “Diabetes is among the commonest causes of chronic morbidity worldwide.” Please rate the completeness of referencing: for most or all relevant key statements (2), inconsistently (1), sporadically (0).

Item 5 – Scientific reasoning

The item describes the quality of the scientific point made. A convincing narrative review presents evidence for key arguments. It should mention study design (randomized controlled trial, qualitative study, etc), and where available, levels of evidence. Please rate whether you feel this has been done thoroughly (2), superficially (1), or hardly at all (0). Unlike item 6, which is concerned with the selection and presentation of concrete outcome data, this item relates to the use of evidence and of types of evidence in the manuscript's arguments.

Item 6 – Appropriate presentation of data:

This item describes the correct presentation of data central to the article's argument. Which data are considered relevant varies from field to field. In some areas relevant data would be absolute rather than relative risks or clinical versus surrogate or intermediate end-points. These outcomes must be presented correctly. For example, it is appropriate that effect sizes are accompanied by confidence intervals. Please rate how far the paper achieves this – thoroughly (2), partially (1), or hardly at all (0). Unlike item 5, which relates to the use of evidence and of types of evidence in the manuscript's arguments, this item is concerned with the selection and presentation of concrete outcome data.

Item 1 – Justification of the article’s importance for the readership

- Página 1: “Acute and chronic poisoning resulting from exposure to heavy metals are uncommon but represent significant morbidity and mortality if unrecognized [1]. Hereupon, clinicians should be aware of the elements that can cause significant harm to patients if not detected and treated quickly.
- Página 2: “ Some of these characteristic signs may lead to the suspicion of a specific metal, and consequently to the early detection of the underlying intoxication as well as the early formulation of emergent treatment.”
- Página 4: “ Even considering that most acute arsenic poisoning occurrences are due to accidental ingestion of arsenic containing pesticides and less commonly from attempted suicide or homicide [12], arsenic is still often seen as the most common homicidal agent. The historical use as a poison has earned it the title of “Poison of Kings and the King of Poisons” [13].”
- Página 8: “Nevertheless, as fluoride has become substantially more used in dental products and was included in food sources (via fluoridated water), several sources of fluoride exposure are now related to the mentioned increase in the incidence of dental fluorosis. Dental fluorosis is reported to be the greatest risk of community water fluoridation [107, 108].”
- Página 18: “The toxicity resulting from intentional or accidental ingestion is a common poisoning. Most of the cases of iron poisoning reported are of pediatric age group, usually in the context of unintentional ingestion [270-272] since iron tablets appear as chocolate candy. Life-threatening toxicity is associated with pediatric ingestion of potent adult preparations, such as prenatal vitamins [273, 274]”
- Página 21: “Furthermore, during recent years, new data suggests that relatively low cadmium exposure, is enough to give rise to skeletal damage, evidenced by osteoporosis and fractures [344-346].”

Item 2: Statement of concrete/specific aims or formulation of questions

- Página 1: “This manuscript aims to collect the clinical and forensic signs related to the poisoning by heavy metals such as thallium, lead, copper, mercury, iron, cadmium and bismuth, as well as other elements such as arsenic, selenium and fluor. Furthermore, their main sources of occupational and environmental exposure are highlighted in this review. The importance of rapid recognition is related to the fact that, through a high degree of suspicion, the clinician could start a treatment, even before the toxicological results are available, which can make a huge difference in these patients outcomes.”

Item 3 – Description of the literature search

- Página 2: “The electronic search took place in PubMed and Google Scholar concerning signs and symptoms, pathophysiology, history and physical examination, diagnostic and forensic signs of intoxication of heavy metals. The keyword “intoxication” was crossed with thallium, arsenic, lead, fluor, copper, selenium, mercury, iron, cadmium, and bismuth as well as their respective forensic and clinical signs. Furthermore, retrieved journal articles, governmental documents and books were reviewed to expand the sources of information. Scientific documents, including books, articles, and government documents, were included in this review, making a total of 401 citations.”

Item 4 – Referencing

- Página 3: “Acute hair loss seems to occur due to the binding between thallium and the cysteine sulfhydryl groups found in hair while dermatitis. Mees’ lines and neuropathy are likely effects of secondary riboflavin deficiency [15, 18, 27, 28].”
- Página 6: “Industrial exposure is primarily via the respiratory tract. Chronic poisoning less commonly results from ingestion [7, 8] although, in pediatric plumbism, pica is usually the common denominator [78-80].”
- Página 8: “Like dental fluorosis, skeleton fluorosis is caused by high levels of fluoride via ingestion or inhalation, over a chronic period of time. High fluoride accumulation in bones and joints creates an imbalance in the bone mineral metabolism resulting in bone reabsorption and abnormal levels of calcium [121-123].”
- Página 10: “Wilson Disease (WD) is a congenital autosomal disorder of copper metabolism produced by a mutation in the ATP 7B copper transporter gene and has a ratio of around 1/30,000-1/100,000 [140]. The copper-transporting P-type ATPase is encoded by the causative gene, ATP7B [141-143]. Mutations that result in a full absence or nonfunctional of ATP7B protein activity are related to early-onset, typically hepatic, severe Wilson’s disease. In comparison, these mutations are rarer [144, 145].”
- Página 11: “The presence of SC turned out to have a limited impact on the patient’s visual acuity. This should be highlighted, as classical cataracts frequently have a marked impairment on visual acuity. SC is not a “true” cataract, as it results from reversible copper deposition below the anterior lens capsule [153, 155, 160-162]. ”
- Página 13: “The typical garlic odor of breath and sweat is the consequence of the production of dimethyl selenide and reflects high blood concentrations of selenium [192,197]. More serious presentations include musculoskeletal dysfunction, neuropathy, liver failure, coma and death [178, 197, 198, 200, 201].”
- Página 19: “The most common and early symptoms include fatigue (70% to 80%), arthralgia/arthritis (40% to 50%), abdominal pain (20% to 60%), decreased libido or impotence (20% to 50%) and weight loss (10% to 50%). The most frequent clinical symptoms for diagnosis are hepatomegaly (50% to 90%), skin hyperpigmentation- a bronze or slate-grey coloration, (30% to 80%), hypogonadism (20% to 50%), arthropathy, splenomegaly, diabetes mellitus, cirrhosis, cardiomyopathy and arrhythmia [295, 304-310].”

Item 5 – Scientific reasoning

- Página 3: “In a study by *Rattner at al.* [64], for an exposure of 4.75 mg/day, hyperpigmentation appeared after six months and hyperkeratosis occurred 3 years after.”
- Página 8: “Dental fluorosis, first described by Trendley Dean in 1937, is a developmental condition of enamel as a result of excessive absorption and repeated exposure to small doses of fluoride during all stages of tooth development [112, 113]. The gravity of fluorosis observed is multifactorial but is deeply connected with both the amount and timing of fluoride exposure [114, 115].”
- Página 10: “These rings are one of the few diagnostic indicators in clinical medicine. Kayser described the rings for the first time in 1902 and then Fleischer did it in 1909 [147, 148]. In 1949, Gerlach and Rohrschneider finally established the fact that the rings were made of copper [149].”
- Página 10: “In 1922, Siemerling and Oloff described an uncommon ocular manifestation of Wilson’s disease known as “sunflower cataract” (Fig. 5F, 5G and 5H). They identified

similarities between the lens changes seen in a patient with Wilson's disease and those caused by an intraocular copper-containing foreign body [153]. Although quite rare [154], the sunflower cataract is a feature of WD and was initially referred to as a pathognomonic sign for WD almost 90 years ago [155, 156]."

- Página 10: "As Duke-Elder described in 1954, SC consists of a thin, centralized opacification that is placed directly below the anterior capsule and encompasses between one-third and one-half of the anterior lens pole surface area. In all situations, the opacification is central and surrounded by other opacifications disposed in ray-like structures around it."
- Página 14: "In 1713, Ramazzini, who is known to be the Father of Industrial Medicine, wrote the renowned treatise "Diseases of Workers," in which he portrayed the toxic manifestations of mercury on surgeons who were treating patients with syphilis by administering mercury inunctions."
- Página 18: "The toxicity resulting from intentional or accidental ingestion is a common poisoning. Most of the cases of iron poisoning reported are of pediatric age group, usually in the context of unintentional ingestion [270-272] since iron tablets appear as chocolate candy."
- Página 20: "Its biohazardous potential was first described in 1817, by Friedrich Stromeyer, in those working in zinc smelters. In 1948, emphysema and proteinuria in industrial workers exposed to Cd dust was described by Friberg, linking cadmium exposure to renal toxicity [317]."
- Página 21: "The association between cadmium and skeletal damage was first described in 1950, in Japan, where the exposure was caused by cadmium- contaminated water used for irrigation of local fields [319, 341]. This disease became known as Itai-Itai (...)"

Item 6 - Appropriate presentation of data

- Página 4 : "In a study by *Rattner et al.* [64], for an exposure of 4.75 mg/day, hyperpigmentation appeared after six months and hyperkeratosis occurred 3 years after."
- Página 6: "Any trail of lead in the human body can be seen as contamination since there is no known physiological role. The United States Centers for Disease Control and Prevention increased, in 2012, the average level of lead in the blood for adults to 10 µg/dL and for children to 5 µg/dL [85]."
- Página 7: "In the skeleton, fluor has a half-life of around 7 years. About 50% of the fluoride becomes incorporated in the hydroxyapatite crystals, by replacing the hydroxide ions, thus altering the structure and size of the crystals. Furthermore, fluoride seems to influence bone turnover through the influence in factors like Runx2 and RANKL [99]."
- Página 8: "Fluoride can be present in an ionized form in groundwaters, and in areas where the soil has a low level of calcium; like regions with high levels of granite or gneiss. In these areas, relatively high fluoride levels are detected in groundwater. When the fluoride level is higher than 1.5 mg/l (1.5 ppm) in drinking water, dental and skeletal fluorosis can happen [111]."
- Página 10: "Wilson Disease (WD) is a congenital autosomal disorder of copper metabolism produced by a mutation in the ATP 7B copper transporter gene and has a ratio of around 1/30,000-1/100,000 [140]. The copper-transporting P-type ATPase is encoded by the causative gene, ATP7B [141-143]. Mutations that result in a full absence or nonfunctional of ATP7B protein activity are related to early-onset, typically hepatic, severe Wilson's disease. In comparison, these mutations are rarer [144, 145]."
- Página 10: "The Kayser-Fleischer ring occurs in more than 99% of patients with neuropsychiatric impairment, but barely in 25-50% of patients with liver disease or a pre-

symptomatic state. The ring usually vanishes with treatment and its continuity or return imply a non-controlled copper state [150-152]."

- Página 12: "Of all the elements, selenium has one of the tightest ranges between dietary insufficiency (<40ug/day) and toxic levels (400 mg/ day)[171], which makes it vital to carefully control intakes by humans and other creature. The suggested dietary remittance is 55 µg/d for persons 14 years or older [172, 173]."
- Página 16: "The FDA propose an acceptable daily intake of 0.4 µg/kg body weight per day [258]. Due to its solubility, approximately 90% of organic mercury is absorbed in the GI tract [259]. It promptly crosses the blood-brain barrier and placenta reaching levels in the brain three to six times those in the blood [260]."
- Página 17: "When measured at its peak, levels below 350 micrograms/dL, between 350 to 500 micrograms/dL and above 500 micrograms/dL are associated with minimal, moderate and severe systemic toxicity, respectively [279]."
- Página 19: "The most common and early symptoms include fatigue (70% to 80%), arthralgia/arthritis (40% to 50%), abdominal pain (20% to 60%), decreased libido or impotence (20% to 50%) and weight loss (10% to 50%). The most frequent clinical symptoms for diagnosis are hepatomegaly (50% to 90%), skin hyperpigmentation (Fig. 8A) - a bronze or slate-grey coloration (30% to 80%), hypogonadism (20% to 50%), arthropathy, splenomegaly, diabetes mellitus, cirrhosis, cardiomyopathy and arrhythmia [295, 304-310]."

2. Normas da revista “Journal of Clinical Medicine”:

Manuscript Submission Overview

Types of Publications

JCM has no restrictions on the length of manuscripts, provided that the text is concise and comprehensive. Full experimental details must be provided so that the results can be reproduced. JCM requires that authors publish all experimental controls and make full datasets available where possible (see the guidelines on [Supplementary Materials](#) and references to unpublished data).

Manuscripts submitted to JCM should neither be published previously nor be under consideration for publication in another journal. The main article types are as follows:

- **Articles:** Original research manuscripts. The journal considers all original research manuscripts provided that the work reports scientifically sound experiments and provides a substantial amount of new information. Authors should not unnecessarily divide their work into several related manuscripts, although Short Communications of preliminary, but significant, results will be considered. The quality and impact of the study will be considered during peer review.
- **Reviews:** These provide concise and precise updates on the latest progress made in a given area of research. Systematic reviews should follow the PRISMA [guidelines](#).

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Manuscripts for JCM should be submitted online at susy.mdpi.com. The submitting author, who is generally the corresponding author, is responsible for the manuscript during the submission and peer-review process. The submitting author must ensure that all eligible co-authors have been included in the author list (read the [criteria to qualify for authorship](#)) and that they have all read and approved the submitted version of the manuscript. To submit your manuscript, register and log in to the [submission website](#). Once you have registered, [click here to go to the submission form for JCM](#). All co-authors can see the manuscript details in the submission system, if they register and log in using the e-mail address provided during manuscript submission.

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Authors must use the [Microsoft Word template](#) or [LaTeX template](#) to prepare their manuscript. Using the template file will substantially shorten the time to complete copy-editing and publication of accepted manuscripts. The total amount of data for all files must not exceed 120 MB. If this is a problem, please contact the Editorial Office jcm@mdpi.com. Accepted file formats are:

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- Your references may be in any style, provided that you use the consistent formatting throughout. It is essential to include author(s) name(s), journal or book title, article or chapter title (where required), year of publication, volume and issue (where appropriate) and pagination. DOI numbers (Digital Object Identifier) are not mandatory but highly encouraged. The bibliography software package EndNote, [Zotero](#), Mendeley, Reference Manager are recommended.

- When your manuscript reaches the revision stage, you will be requested to format the manuscript according to the journal guidelines.

Cover Letter

A cover letter must be included with each manuscript submission. It should be concise and explain why the content of the paper is significant, placing the findings in the context of existing work and why it fits the scope of the journal. Confirm that neither the manuscript nor any parts of its content are currently under consideration or published in another journal. Any prior submissions of the manuscript to MDPI journals must be acknowledged. The names of proposed and excluded reviewers should be provided in the submission system, not in the cover letter.

Author Biography

Authors are encouraged to add a biography (maximum 150 words) to the submission and publish it. This should be a single paragraph and should contain the following points:

1. Authors' full names followed by current positions;
2. Education background including institution information and year of graduation (type and level of degree received);
3. Work experience;
4. Current and previous research interests;
5. Memberships of professional societies and awards received.

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This journal automatically deposits papers to PubMed Central after publication of an issue. Authors do not need to separately submit their papers through the NIH Manuscript Submission System (NIHMS, <http://nihms.nih.gov/>).

Registered Reports

Registered Reports are scientific articles which are peer-reviewed before the research is performed and the data are collected.

The ideas that meet high scientific standards, such as rigor, soundness, significant importance and implications for the science community, are then provisionally accepted for publication before data collection starts.

This format helps to considerably reduce publication bias and to avoid researchers having to invest much time and effort into questionable research studies, while allowing enough flexibility to perform unregistered experiments and report serendipitous findings.

The review process for Registered Reports can be briefly summarize into two main stages.

Stage 1:

- Submission: Authors propose a clear hypothesis (including background, experimental methods and analyses) that they plan to investigate.
- Peer review: Reviewers evaluate the registered study proposal before data are collected (research importance, rigor, soundness, etc.)

Following in-principle acceptance (IPA), the authors are asked to perform the experiments, following exactly the peer-reviewed procedures.

Stage 2:

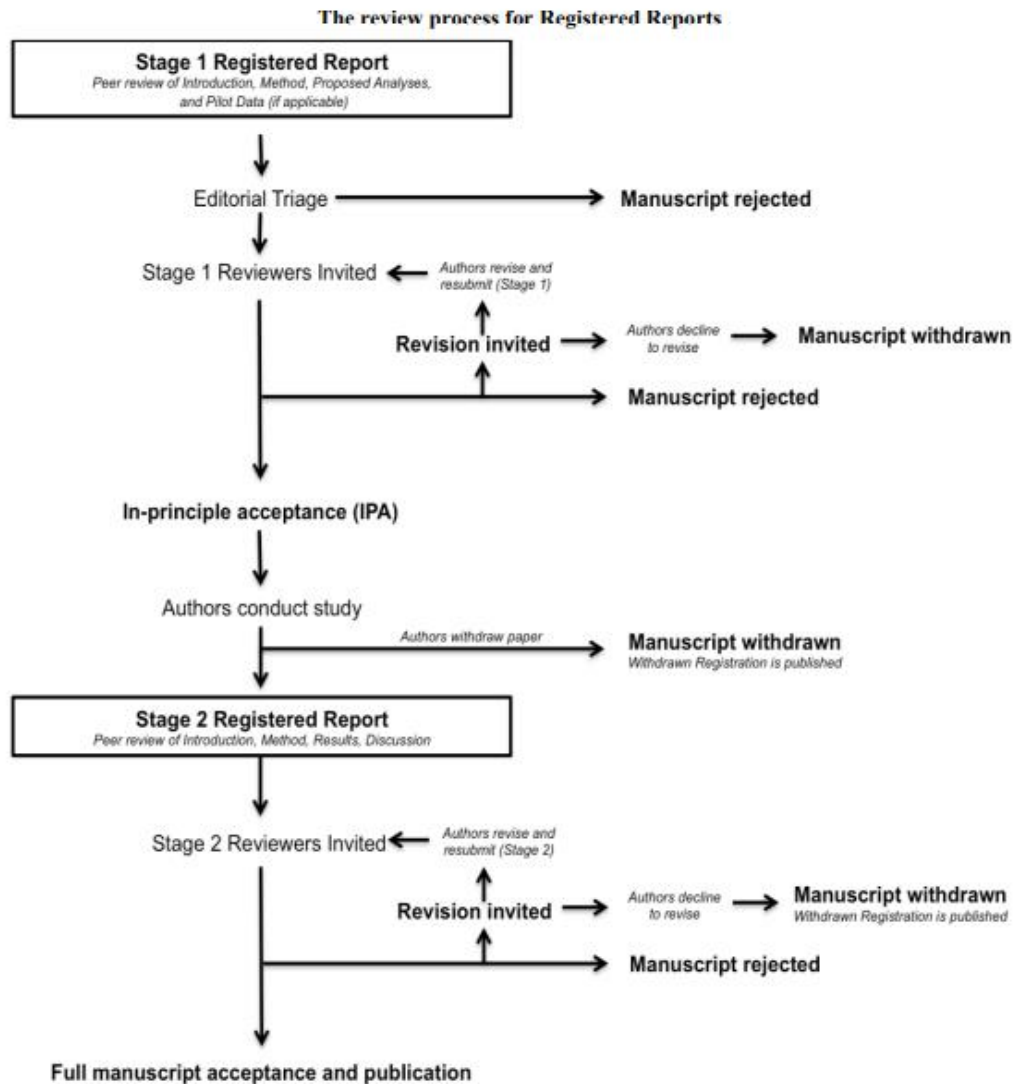
- Submission: Authors come back with a complete study, including the results and analyses.
- Peer review: Reviewers take another look, this time considering the full study and assessing compliance with the original protocol.

Guidelines for Authors

Registered Reports are a form of empirical article in which the methods and proposed analyses are pre-registered and reviewed prior to research being conducted. This format is designed to minimise bias in deductive science, while also allowing complete flexibility to conduct exploratory (unregistered) analyses and report serendipitous findings.

The cornerstone of the Registered Reports format is that a significant part of the manuscript will be assessed prior to data collection, with the highest quality submissions accepted in advance. Initial submissions will include a description of the key research question and background literature, hypotheses, experimental procedures, analysis pipeline, a statistical power analysis (or Bayesian equivalent), and pilot data (where applicable).

Initial submissions will be triaged by an editorial team for suitability. Those that pass triage will then be sent for in-depth peer review (Stage 1). Following review, the article will then be either rejected or accepted in principle for publication. Following in principle acceptance (IPA), the authors will then proceed to conduct the study, adhering exactly to the peer-reviewed procedures. When the study is complete the authors will submit their finalised manuscript for re-review (Stage 2) and will upload their raw data, digital study materials, and laboratory log to a publicly accessible file-sharing service. Pending quality checks and a sensible interpretation of the findings, the manuscript will be published regardless of the results.



Stage 1: Initial manuscript submission and review

Stage 1 submissions should include the manuscript (details below) and a brief cover letter. Please note that the editorial board will not agree to send manuscripts for in-depth review until a complete Stage 1 submission has been considered.

The title of the paper should include words “Stage 1”.

The Stage 1 cover letter should include:

- A brief scientific case for consideration. Authors are encouraged to refer to the likely replication value of the research. Replication studies are welcome in addition to novel studies.
- A statement confirming that all necessary support (e.g., funding, facilities) and approvals (e.g., ethics) are in place for the proposed research. Note that manuscripts will be generally considered only for studies that are able to commence immediately; however, authors with alternative plans are encouraged to contact the journal office for advice.
- An anticipated timeline (typically within 12 months) for completing the study if the initial submission is accepted. Authors should contact the Editorial team for advice if longer time is needed.

- A statement confirming that the authors agree to share their raw data, any digital study materials, and analysis code as appropriate.
- A statement confirming that, following Stage 1 in principle acceptance, the authors choose to publish Stage 1 papers in the journal.

Manuscript preparation guidelines – Stage 1

Initial Stage 1 submissions should include the following sections

- Introduction
 - o A review of the relevant literature that motivates the research question and a full description of the experimental aims and hypotheses. Please note that following IPA, the Introduction section cannot be altered apart from correction of factual errors, typographic errors and altering of tense from future to past (see below).
- Methods
 - o Full description of proposed sample characteristics, including criteria for data inclusion and exclusion (e.g., outlier extraction). Procedures for objectively defining exclusion criteria due to technical errors or for any other reasons must be specified, including details of how and under what conditions data would be replaced.
 - o A description of experimental procedures in sufficient detail to allow another researcher to repeat the methodology exactly, without requiring further information. These procedures must be adhered to exactly in the subsequent experiments or any Stage 2 manuscript can be rejected.
 - o Proposed analysis pipeline, including all preprocessing steps, and a precise description of all planned analyses, including appropriate correction for multiple comparisons. Any covariates or regressors must be stated. Where analysis decisions are contingent on the outcome of prior analyses, these contingencies must be specified and adhered to. Only pre-planned analyses can be reported in the main Results section of Stage 2 submissions. However, unplanned exploratory analyses will be admissible in a separate section of the Results (see below).
 - o Studies involving Neyman-Pearson inference must include a statistical power analysis. Estimated effect sizes should be justified with reference to the existing literature or theory. Since publication bias overinflates published estimates of effect size, power analysis must be based on the lowest available or meaningful estimate of the effect size. For frequentist analysis plans, the a priori power must be 0.9 or higher for all proposed hypothesis tests. In the case of highly uncertain effect sizes, a variable sample size and interim data analysis is permissible but with inspection points stated in advance, appropriate Type I error correction for ‘peeking’ employed, and a final stopping rule for data collection outlined.
 - o Methods involving Bayesian hypothesis testing are encouraged. For studies involving analyses with Bayes factors, the predictions of the theory must be specified so that a Bayes factor can be calculated. Authors should indicate what distribution will be used to represent the predictions of the theory and how its parameters will be specified. For example, will you use a uniform up to some specified maximum, or a normal/half-normal to represent a likely effect size, or a JZS/Cauchy with a specified scaling constant? For inference by Bayes factors, authors must be able to guarantee data collection until the Bayes factor is at least 6 times in favour of the experimental hypothesis over the null hypothesis (or vice versa). Authors with resource limitations are permitted to specify a maximum feasible sample size at which data collection must

cease regardless of the Bayes factor; however to be eligible for advance acceptance this number must be sufficiently large that inconclusive results at this sample size would nevertheless be an important message for the field. For further advice on Bayes factors or Bayesian sampling methods, prospective authors are encouraged to read this key article by Schönbrodt and Wagenmakers.

- o Full descriptions must be provided of any outcome-neutral criteria that must be met for successful testing of the stated hypotheses. Such quality checks might include the absence of floor or ceiling effects in data distributions, positive controls, or other quality checks that are orthogonal to the experimental hypotheses.

- o A timeline for completion of the study and proposed resubmission date if Stage 1 review is successful must be included. Extensions to this deadline can be negotiated with the Editorial Team.

- o Any description of prospective methods or analysis plans should be written in future tense.

- Pilot Data

- o Optional. Can be included to establish proof of concept, effect size estimations, or feasibility of proposed methods. Any pilot experiments will be published with the final version of the manuscript and will be clearly distinguished from data obtained for the pre-registered experiment(s).

- Secondary Registrations

- o The journal welcomes submissions proposing secondary analyses of existing data sets, provided authors can supply sufficient evidence (e.g., self-certification; letter from independent gatekeeper) to confirm that they have had no prior access to the data in question. For advice on the eligibility of specific scenarios, authors are welcome to contact the editorial office.

Stage 1 submissions that are judged by the editorial board to be of sufficient quality and within journal scope will be sent for indepth peer review. In considering papers at the registration stage, reviewers will be asked to assess:

1. The importance and soundness of the proposed hypotheses
2. The suitability and feasibility of the experimental and analysis methodology
3. Whether there are sufficient details given to replicate the proposed experimental procedures and analysis
4. Whether there are sufficient outcome-neutral tests of the hypotheses, including positive controls and quality checks

Following Stage 1 peer review, manuscripts will be rejected outright, offered the opportunity to revise, or accepted. Proposals that exceed the highest standards of importance and scientific rigour will be issued an in principle acceptance (IPA), indicating that the article will be published pending completion of the approved methods and analytic procedures, passing of all pre-specified quality checks, and a defensible interpretation of the results. Manuscripts that pass Stage 1 peer review will be published immediately. Authors will be required to cite Stage 1 papers in the final Stage 2 manuscripts and link it to Stage 1, indicating the manuscript is a registered report.

Authors are reminded that any deviation from the stated experimental procedures, regardless of how minor it may seem to the authors, could lead to rejection of the manuscript at Stage 2.

In cases where the pre-registered protocol is altered after IPA due to unforeseen circumstances (e.g., change of equipment or unanticipated technical error), the authors must consult the editorial team immediately for advice, and prior to the completion of data collection. Minor changes to the protocol may be permitted per editorial discretion. In such cases, IPA would be preserved and the deviation reported in the Stage 2 submission. Note that registered analyses must be undertaken, but additional unregistered analyses can also be included in a final manuscript (see below).

Stage 2: Full manuscript review

Once the study is complete, authors prepare and resubmit their manuscript for full review, with the following additions:

- The title of the paper should include words “Stage 2”.
- Cover letter. The Stage 2 cover letter must confirm:
 - o That the manuscript includes a link to the published Stage 1 paper, or to the public archive containing anonymized study data, digital materials/code and the laboratory log. The cover letter should state the page number in the manuscript that lists the URL.
 - o That, for primary Registered Reports, no data for any pre-registered study (other than pilot data included at Stage 1) was collected prior to the date of IPA. For secondary Registered Reports, authors should confirm that no data (other than pilot data included at Stage 1) was subjected to the pre-registered analyses prior to IPA.
- Submission of anonymised raw data, digital study materials, and laboratory log
 - o Anonymised raw data and digital study materials must be made freely available in a public repository/archive with a link provided within the Stage 2 manuscript. Authors are free to use any repository that renders data and materials freely and publicly accessible and provides a digital object identifier (DOI) to ensure that the data remain persistent, unique and citable. Potential repositories include (but are not limited to), Figshare, Harvard Dataverse, and Dryad. For a comprehensive list of available data repositories, see <http://www.re3data.org/>.
 - o Data files should be appropriately time stamped to show that data was collected after IPA and not before. Other than pre-registered and approved pilot data, no data acquired prior to the date of IPA is admissible in the Stage 2 submission. Raw data must be accompanied by guidance notes, where required, to assist other scientists in replicating the analysis pipeline. Authors are required to upload any relevant analysis scripts and other digital experimental materials that would assist in replication.
 - o Any supplementary figures, tables, or other text (such as supplementary methods) can either be included as standard supplementary information that accompanies the paper, or they can be archived together with the data. Please note that the raw data itself should be archived (see above) rather than submitted to the journal as supplementary material.
 - o A basic laboratory log must also be provided outlining the range of dates during which data collection took place. This log should be uploaded to the same public archive as the data and materials.

- Background, Rationale and Methods

- o Apart from minor stylistic revisions, the Introduction cannot be altered from the approved Stage 1 submission, and the stated hypotheses cannot be amended or appended. At Stage 2, any description of the rationale or proposed methodology that was written in future tense within the Stage 1 manuscript should be changed to past tense. Any textual changes to the Introduction or Methods (e.g. correction of typographic errors) must be clearly marked in the Stage 2 submission. Any relevant literature that appeared following the date of IPA should be covered in the Discussion.

- o The outcome of all registered analyses must be reported in the manuscript, except in rare instances where a registered and approved analysis is subsequently shown to be logically flawed or unfounded. In such cases, the authors, reviewers, and editor must agree that a collective error of judgment was made and that the analysis is inappropriate. In such cases the analysis would still be mentioned in the Methods but omitted with justification from the Results.

- o It is reasonable that authors may wish to include additional analyses that were not included in the registered submission. For instance, a new analytic approach might become available between IPA and Stage 2 review, or a particularly interesting and unexpected finding may emerge. Such analyses are admissible but must be clearly justified in the text, appropriately caveated, and reported in a separate section of the Results titled “Exploratory Analyses”. Authors should be careful not to base their conclusions entirely on the outcome of statistically significant post hoc analyses.

- o Authors reporting null hypothesis significance tests are required to report exact p values and effect sizes for all inferential analyses.

The resubmission will most likely be considered by the same reviewers as in Stage 1, but could also be assessed by new reviewers. In considering papers at Stage 2, reviewers will be asked to decide:

1. Whether the data are able to test the original proposed hypotheses by satisfying the approved outcome-neutral conditions (such as quality checks, positive controls)
2. Whether the stated hypotheses tested was the same as the approved Stage 1 submission
3. Whether the authors adhered precisely to the registered experimental procedures or were able to sufficiently justify any changes
4. Whether any new analyses (not mentioned at Stage 1) are methodologically sound and relevant
5. Whether the authors’ conclusions are justified given the data

Reviewers are informed that editorial decisions will not be based on the perceived importance, novelty or conclusiveness of the results. Thus, while reviewers are free to enter such comments on the record, they will not influence editorial decisions. Reviewers at Stage 2 may suggest that authors report additional post hoc tests on their data; however, authors are not obliged to do so unless such tests are necessary to satisfy one or more of the Stage 2 review criteria.

Manuscript withdrawal

It is possible that authors with IPA may wish to withdraw their manuscript following or during data collection. Possible reasons could include major technical error, an inability to complete the study due to other unforeseen circumstances, or the desire to submit the results to a different journal. In all such cases, manuscripts can of course be withdrawn at the authors’ discretion.

Incremental Registrations

Authors may add experiments to approved submissions. In such cases the approved Stage 2 manuscript will be accepted for publication, and authors can propose additional experiments for Stage 1

consideration. Where these experiments extend the approved submission (as opposed to being part of new submissions), the editorial team will seek to fast-track the review process. This option may be particularly appropriate where an initial experiment reveals a major serendipitous finding that warrants followup within the same paper. In cases where an incremented submission is rejected (at either Stage 1 or 2), authors will retain the option of publishing the most recently approved version of the manuscript. For further advice on specific scenarios for incremental registration, authors are invited to contact the editorial office.

Tips for Avoiding Desk Rejection at Stage 1

Many Registered Report submissions are desk rejected at Stage 1, prior to in-depth review, for failing to sufficiently meet the Stage 1 editorial criteria. In many such cases, authors are invited to resubmit once specific shortcomings are addressed, although major problems can lead to outright rejection. To help minimize the chances of authors' submissions being desk rejected, we list below the top ten reasons why Stage 1 submissions are rejected prior to review.

1. Cover letter doesn't make necessary statements concerning ethics, data archiving, and so forth (see above).

2. The protocol contains insufficient methodological detail to enable replication and prevent researcher degrees of freedom. One commonly neglected area is the criteria for excluding data, both at the level of animals/participants and at the level of data within animals/participants. In the interests of clarity, we recommend listing these criteria systematically rather than presenting them in prose.

3. Lack of correspondence between the scientific hypotheses and the pre-registered statistical tests. This is a common problem and severe cases are likely to be desk rejected outright. To maximize clarity of correspondence between predictions and analyses, authors are encouraged to number their hypotheses in the Introduction and then number the proposed analyses in the Methods to make clear which analysis tests which prediction. Ensure also that power analysis, where applicable, is based on the actual test procedures that will be employed to test those hypotheses; e.g. don't propose a power analysis based on an ANOVA but then suggest a linear mixed effects model to test the hypothesis.

4. Power analysis, where applicable, fails to reach the minimum level stated in journal policy

5. Power analysis is over-optimistic (e.g. based on previous literature but not taking into account publication bias) or insufficiently justified (e.g. based on a single point estimate from a pilot experiment or previous study). Proposals should be powered to detect the smallest effect that is plausible and of theoretical value. Pilot data can help inform this estimate but is unlikely to form an acceptable basis, alone, for choosing the target effect size.

6. Intention to infer support for the null hypothesis from statistically non-significant results, without proposing use of Bayes factors or frequentist equivalence testing.

7. Inclusion of exploratory analyses in the analysis plan. Manuscripts proposing exploratory analyses will usually be desk rejected until such analyses are removed because inclusion of exploratory "plans" at Stage 1 blurs the line between confirmatory and exploratory outcomes at Stage 2. Instead, such analyses can be included at Stage 2 and need not be preregistered. Under some circumstances, exploratory analyses could be discussed at Stage 1 where they are necessary to justify study variables or procedures that are included in the design exclusively for exploratory analysis.

8. Failure to clearly distinguish work that has already been done from work that is planned. Where a proposal contains a mixture of pilot work that has already been undertaken and a proposal for work not yet undertaken, authors should use the past tense for pilot work but the future tense for the proposed work. At Stage 2, all descriptions shift to past tense.

9. Lack of pre-specified positive controls or other quality checks, or an appropriate justification for their absence (See Stage 1 criterion 5). We recognise that positive controls are not possible with all study designs, in which case authors should discuss why they are not included.

10. Where applicable, lack of power analysis within proposed positive controls that depend on hypothesis testing.

Manuscript Preparation

General Considerations

- **Research manuscripts** should comprise:

- o Front matter: Title, Author list, Affiliations, Abstract, Keywords

- o Research manuscript sections: Introduction, Materials and Methods, Results, Discussion, Conclusions (optional).

- o Back matter: Supplementary Materials, Acknowledgments, Author Contributions, Conflicts of Interest, References.

- **Review manuscripts** should comprise the front matter, literature review sections and the back matter. The template file can also be used to prepare the front and back matter of your review manuscript. It is not necessary to follow the remaining structure. Structured reviews and meta-analyses should use the same structure as research articles and ensure they conform to the PRISMA guidelines.

- **Graphical Abstract:**

A graphical abstract (GA) is an image that appears alongside the text abstract in the Table of Contents. In addition to summarizing content, it should represent the topic of the article in an attention-grabbing way.

The GA should be a high-quality illustration or diagram in any of the following formats: PNG, JPEG, EPS, SVG, PSD or AI. Written text in a GA should be clear and easy to read, using one of the following fonts: Times, Arial, Courier, Helvetica, Ubuntu or Calibri.

The minimum required size for the GA is 560 × 1100 pixels (height × width). When submitting larger images, please make sure to keep to the same ratio.

- **Abbreviations** should be defined in parentheses the first time they appear in the abstract, main text, and in figure or table captions and used consistently thereafter.

- **SI Units** (International System of Units) should be used. Imperial, US customary and other units should be converted to SI units whenever possible.

- **Accession numbers** of RNA, DNA and protein sequences used in the manuscript should be provided in the Materials and Methods section. Also see the section on Deposition of Sequences and of Expression Data.

- **Equations:** If you are using Word, please use either the Microsoft Equation Editor or the MathType add-on. Equations should be editable by the editorial office and not appear in a picture format.

- **Research Data and supplementary materials:** Note that publication of your manuscript implies that you must make all materials, data, and protocols associated with the publication available to readers. Disclose at the submission stage any restrictions on the availability of materials or information. Read the information about Supplementary Materials and Data Deposit for additional guidelines.

- **Preregistration:** Where authors have preregistered studies or analysis plans, links to the preregistration must be provided in the manuscript.
- **Guidelines and standards:** MDPI follows standards and guidelines for certain types of research. See https://www.mdpi.com/editorial_process for further information.

Front Matter

These sections should appear in all manuscript types

- **Title:** The title of your manuscript should be concise, specific and relevant. It should identify if the study reports (human or animal) trial data, or is a systematic review, meta-analysis or replication study. When gene or protein names are included, the abbreviated name rather than full name should be used.
- **Author List and Affiliations:** Authors' full first and last names must be provided. The initials of any middle names can be added. The PubMed/MEDLINE standard format is used for affiliations: complete address information including city, zip code, state/province, and country. At least one author should be designated as corresponding author, and his or her email address and other details should be included at the end of the affiliation section. Please read the criteria to qualify for authorship.
- **Abstract:** The abstract should be a total of about 200 words maximum. The abstract should be a single paragraph and should follow the style of structured abstracts, but without headings: 1) Background: Place the question addressed in a broad context and highlight the purpose of the study; 2) Methods: Describe briefly the main methods or treatments applied. Include any relevant preregistration numbers, and species and strains of any animals used. 3) Results: Summarize the article's main findings; and 4) Conclusion: Indicate the main conclusions or interpretations. The abstract should be an objective representation of the article: it must not contain results which are not presented and substantiated in the main text and should not exaggerate the main conclusions.
- **Keywords:** Three to ten pertinent keywords need to be added after the abstract. We recommend that the keywords are specific to the article, yet reasonably common within the subject discipline.

Research Manuscript Sections

- **Introduction:** The introduction should briefly place the study in a broad context and highlight why it is important. It should define the purpose of the work and its significance, including specific hypotheses being tested. The current state of the research field should be reviewed carefully and key publications cited. Please highlight controversial and diverging hypotheses when necessary. Finally, briefly mention the main aim of the work and highlight the main conclusions. Keep the introduction comprehensible to scientists working outside the topic of the paper.
- **Materials and Methods:** They should be described with sufficient detail to allow others to replicate and build on published results. New methods and protocols should be described in detail while well-established methods can be briefly described and appropriately cited. Give the name and version of any software used and make clear whether computer code used is available. Include any pre-registration codes.
- **Results:** Provide a concise and precise description of the experimental results, their interpretation as well as the experimental conclusions that can be drawn.
- **Discussion:** Authors should discuss the results and how they can be interpreted in perspective of previous studies and of the working hypotheses. The findings and their implications should be

discussed in the broadest context possible and limitations of the work highlighted. Future research directions may also be mentioned. This section may be combined with Results.

- **Conclusions:** This section is not mandatory, but can be added to the manuscript if the discussion is unusually long or complex.
- **Patents:** This section is not mandatory, but may be added if there are patents resulting from the work reported in this manuscript.

Back Matter

- **Supplementary Materials:** Describe any supplementary material published online alongside the manuscript (figure, tables, video, spreadsheets, etc.). Please indicate the name and title of each element as follows Figure S1: title, Table S1: title, etc.

- **Funding:** All sources of funding of the study should be disclosed. Clearly indicate grants that you have received in support of your research work and if you received funds to cover publication costs. Note that some funders will not refund article processing charges (APC) if the funder and grant number are not clearly and correctly identified in the paper. Funding information can be entered separately into the submission system by the authors during submission of their manuscript. Such funding information, if available, will be deposited to FundRef if the manuscript is finally published. Please add: "This research received no external funding" or "This research was funded by [name of funder] grant number [xxx]" and "The APC was funded by [XXX]" in this section. Check carefully that the details given are accurate and use the standard spelling of funding agency names at <https://search.crossref.org/funding>, any errors may affect your future funding.

- **Acknowledgments:** In this section you can acknowledge any support given which is not covered by the author contribution or funding sections. This may include administrative and technical support, or donations in kind (e.g., materials used for experiments).

- **Author Contributions:** Each author is expected to have made substantial contributions to the conception or design of the work; or the acquisition, analysis, or interpretation of data; or the creation of new software used in the work; or have drafted the work or substantively revised it; AND has approved the submitted version (and version substantially edited by journal staff that involves the author's contribution to the study); AND agrees to be personally accountable for the author's own contributions and for ensuring that questions related to the accuracy or integrity of any part of the work, even ones in which the author was not personally involved, are appropriately investigated, resolved, and documented in the literature. For research articles with several authors, a short paragraph specifying their individual contributions must be provided. The following statements should be used "Conceptualization, X.X. and Y.Y.; Methodology, X.X.; Software, X.X.; Validation, X.X., Y.Y. and Z.Z.; Formal Analysis, X.X.; Investigation, X.X.; Resources, X.X.; Data Curation, X.X.; Writing – Original Draft Preparation, X.X.; Writing – Review & Editing, X.X.; Visualization, X.X.; Supervision, X.X.; Project Administration, X.X.; Funding Acquisition, Y.Y.", please turn to the CRediT taxonomy for the term explanation. For more background on CRediT, see here. **Authorship must include and be limited to those who have contributed substantially to the work. Please read the section concerning the [criteria to qualify for authorship](#) carefully**".

- **Institutional Review Board Statement:** In this section, please add the Institutional Review Board Statement and approval number for studies involving humans or animals. Please note that the

Editorial Office might ask you for further information. Please add “The study was conducted according to the guidelines of the Declaration of Helsinki, and approved by the Institutional Review Board (or Ethics Committee) of NAME OF INSTITUTE (protocol code XXX and date of approval).” OR “Ethical review and approval were waived for this study, due to REASON (please provide a detailed justification).” OR “Not applicable” for studies not involving humans or animals. You might also choose to exclude this statement if the study did not involve humans or animals.

- **Informed Consent Statement:** Any research article describing a study involving humans should contain this statement. Please add “Informed consent was obtained from all subjects involved in the study.” OR “Patient consent was waived due to REASON (please provide a detailed justification).” OR “Not applicable” for studies not involving humans. You might also choose to exclude this statement if the study did not involve humans. Written informed consent for publication must be obtained from participating patients who can be identified (including by the patients themselves). Please state “Written informed consent has been obtained from the patient(s) to publish this paper” if applicable.

- **Data Availability Statement:** In this section, please provide details regarding where data supporting reported results can be found, including links to publicly archived datasets analyzed or generated during the study. Please refer to suggested 50 Data Availability Statements in section “[MDPI Research Data Policies](#)”. You might choose to exclude this statement if the study did not report any data.

- **Conflicts of Interest:** Authors must identify and declare any personal circumstances or interest that may be perceived as influencing the representation or interpretation of reported research results. If there is no conflict of interest, please state “The authors declare no conflict of interest.” Any role of the funding sponsors in the choice of research project; design of the study; in the collection, analyses or interpretation of data; in the writing of the manuscript; or in the decision to publish the results must be declared in this section. JCM does not publish studies funded by the tobacco industry. Any projects funded by industry must pay special attention to the full declaration of funder involvement. If there is no role, please state “The sponsors had no role in the design, execution, interpretation, or writing of the study”. For more details please see Conflict of Interest.

- **References:** References must be numbered in order of appearance in the text (including table captions and figure legends) and listed individually at the end of the manuscript. We recommend preparing the references with a bibliography software package, such as EndNote, ReferenceManager or Zotero to avoid typing mistakes and duplicated references. We encourage citations to data, computer code and other citable research material. If available online, you may use reference style 9. below.

- Citations and References in Supplementary files are permitted provided that they also appear in the main text and in the reference list.

In the text, reference numbers should be placed in square brackets [], and placed before the punctuation; for example [1], [1–3] or [1,3]. For embedded citations in the text with pagination, use both parentheses and brackets to indicate the reference number and page numbers; for example [5] (p. 10). or [6] (pp. 101–105).

The reference list should include the full title, as recommended by the ACS style guide. Style files for Endnote and Zotero are available.

References should be described as follows, depending on the type of work:

- **Journal Articles:**

1. Author 1, A.B.; Author 2, C.D. Title of the article. Abbreviated Journal Name Year, Volume, page range.

- **Books and Book Chapters:**

2. Author 1, A.; Author 2, B. Book Title, 3rd ed.; Publisher: Publisher Location, Country, Year; pp. 154–196.

3. Author 1, A.; Author 2, B. Title of the chapter. In Book Title, 2nd ed.; Editor 1, A., Editor 2, B., Eds.; Publisher: Publisher Location, Country, Year; Volume 3, pp. 154–196.

• Unpublished work, submitted work, personal communication:

4. Author 1, A.B.; Author 2, C. Title of Unpublished Work. status (unpublished; manuscript in preparation).

5. Author 1, A.B.; Author 2, C. Title of Unpublished Work. Abbreviated Journal Name stage of publication (under review; accepted; in press).

6. Author 1, A.B. (University, City, State, Country); Author 2, C. (Institute, City, State, Country). Personal communication, Year.

• Conference Proceedings:

7. Author 1, A.B.; Author 2, C.D.; Author 3, E.F. Title of Presentation. In Title of the Collected Work (if available), Proceedings of the Name of the Conference, Location of Conference, Country, Date of Conference; Editor 1, Editor 2, Eds. (if available); Publisher: City, Country, Year (if available); Abstract Number (optional), Pagination (optional).

• Thesis:

8. Author 1, A.B. Title of Thesis. Level of Thesis, Degree-Granting University, Location of University, Date of Completion.

• Websites:

9. Title of Site. Available online: URL (accessed on Day Month Year).

Unlike published works, websites may change over time or disappear, so we encourage you create an archive of the cited website using a service such as WebCite. Archived websites should be cited using the link provided as follows:

10. Title of Site. URL (archived on Day Month Year).

See the Reference List and Citations Guide for more detailed information.

Preparing Figures, Schemes and Tables

• File for Figures and Schemes must be provided during submission in a single zip archive and at a sufficiently high resolution (minimum 1000 pixels width/height, or a resolution of 300 dpi or higher). Common formats are accepted, however, TIFF, JPEG, EPS and PDF are preferred.

• JCM can publish multimedia files in articles or as supplementary materials. Please contact the editorial office for further information.

• All Figures, Schemes and Tables should be inserted into the main text close to their first citation and must be numbered following their number of appearance (Figure 1, Scheme I, Figure 2, Scheme II, Table 1, etc.).

• All Figures, Schemes and Tables should have a short explanatory title and caption.

• All table columns should have an explanatory heading. To facilitate the copy-editing of larger tables, smaller fonts may be used, but no less than 8 pt. in size. Authors should use the Table option of Microsoft Word to create tables.

• Authors are encouraged to prepare figures and schemes in color (RGB at 8-bit per channel). There is no additional cost for publishing full color graphics.

Supplementary Materials, Data Deposit and Software Source Code

MDPI Research Data Policies

MDPI is committed to supporting open scientific exchange and enabling our authors to achieve best practices in sharing and archiving research data. We encourage all authors of articles published in MDPI journals to share their research data. Individual journal guidelines can be found at the journal 'Instructions for Authors' page. Data sharing policies concern the minimal dataset that supports the central findings of a published study. Generated data should be publicly available and cited in accordance with journal guidelines.

MDPI data policies are informed by [TOP Guidelines](#).

Where ethical, legal or privacy issues are present, data should not be shared. The authors should make any limitations clear in the Data Availability Statement upon submission. Authors should ensure that data shared are in accordance with consent provided by participants on the use of confidential data.

Data Availability Statements provide details regarding where data supporting reported results can be found, including links to publicly archived datasets analyzed or generated during the study.

Below are suggested Data Availability Statements:

- Data available in a publicly accessible repository The data presented in this study are openly available in [repository name e.g., FigShare] at [doi], reference number [reference number].
- Data available in a publicly accessible repository that does not issue DOIs Publicly available datasets were analyzed in this study. This data can be found here: [link/accession number]
- Data available on request due to restrictions eg privacy or ethical The data presented in this study are available on request from the corresponding author. The data are not publicly available due to [insert reason here]
- 3rd Party Data Restrictions apply to the availability of these data. Data was obtained from [third party] and are available [from the authors / at URL] with the permission of [third party].
- Data sharing not applicable No new data were created or analyzed in this study. Data sharing is not applicable to this article.
- Data is contained within the article or supplementary material The data presented in this study are available in [insert article or supplementary material here]

Data citation:

- [dataset] Authors. Year. Dataset title; Data repository or archive; Version (if any); Persistent identifier (e.g., DOI).

Computer Code and Software

For work where novel computer code was developed, authors should release the code either by depositing in a recognized, public repository or uploading as supplementary information to the publication. The name and version of all software used should be clearly indicated.

Supplementary Material

Additional data and files can be uploaded as "Supplementary Files" during the manuscript submission process. The supplementary files will also be available to the referees as part of the peer-review process. Any file format is acceptable, however we recommend that common, non-proprietary formats are used where possible.

Unpublished Data

Restrictions on data availability should be noted during submission and in the manuscript. "Data not shown" should be avoided: authors are encouraged to publish all observations related to the submitted manuscript as Supplementary Material. "Unpublished data" intended for publication in a manuscript that is either planned, "in preparation" or "submitted" but not yet accepted, should be cited in the text and a reference should be added in the References section. "Personal Communication" should also be cited in the text and reference added in the References section. (see also the MDPI reference list and citations style guide).

Remote Hosting and Large Data Sets

Data may be deposited with specialized service providers or institutional/subject repositories, preferably those that use the DataCite mechanism. Large data sets and files greater than 60 MB must be deposited in this way. For a list of other repositories specialized in scientific and experimental data, please consult databib.org or re3data.org. The data repository name, link to the data set (URL) and 52 accession number, doi or handle number of the data set must be provided in the paper. The journal Data also accepts submissions of data set papers.

Deposition of Sequences and of Expression Data

New sequence information must be deposited to the appropriate database prior to submission of the manuscript. Accession numbers provided by the database should be included in the submitted manuscript. Manuscripts will not be published until the accession number is provided.

- New nucleic acid sequences must be deposited in one of the following databases: GenBank, EMBL, or DDBJ. Sequences should be submitted to only one database.
- New high throughput sequencing (HTS) datasets (RNA-seq, ChIP-Seq, degradome analysis, ...) must be deposited either in the GEO database or in the NCBI's Sequence Read Archive.
- New microarray data must be deposited either in the GEO or the ArrayExpress databases. The "Minimal Information About a Microarray Experiment" (MIAME) guidelines published by the Microarray Gene Expression Data Society must be followed.
- New protein sequences obtained by protein sequencing must be submitted to UniProt (submission tool SPIN).

All sequence names and the accession numbers provided by the databases should be provided in the Materials and Methods section of the article.

References in Supplementary Files

Citations and References in Supplementary files are permitted provided that they also appear in the reference list of the main text.

Research and Publication Ethics

Research Ethics

Research Involving Human Subjects

When reporting on research that involves human subjects, human material, human tissues, or human data, authors must declare that the investigations were carried out following the rules of the Declaration of Helsinki of 1975 (<https://www.wma.net/what-wedo/medical-ethics/declaration-of-helsinki/>), revised in 2013. According to point 23 of this declaration, an approval from an ethics committee should have been obtained before undertaking the research. At a minimum, a statement including the project identification code, date of approval, and name of the ethics committee or institutional review board should be stated in Section 'Institutional Review Board Statement' of the article. Data relating to individual participants must be described in detail, but private information identifying participants need not be included unless the identifiable materials are of relevance to the research (for example, photographs of participants' faces that show a particular symptom). Editors reserve the right to reject any submission that does not meet these requirements.

Example of an ethical statement: "All subjects gave their informed consent for inclusion before they participated in the study. The study was conducted in accordance with the Declaration of Helsinki, and the protocol was approved by the Ethics Committee of XXX (Project identification code)."

A written informed consent for publication must be obtained from participating patients who can be identified (including by the patients themselves). Patients' initials or other personal identifiers must not appear in any images. For manuscripts that include any case details, personal information, and/or images of patients, authors must obtain signed informed consent from patients (or their relatives/guardians) before submitting to an MDPI journal. Patient details must be anonymized as far as possible, e.g., do not mention specific age, ethnicity, or occupation where they are not relevant to the conclusions. A [template permission form](#) is available to download. A blank version of the form used to obtain permission (without the patient names or signature) must be uploaded with your submission.

You may refer to our sample form and provide an appropriate form after consulting with your affiliated institution. Alternatively, you may provide a detailed justification of why informed consent is not necessary. For the purposes of publishing in MDPI journals, a consent, permission, or release form should include unlimited permission for publication in all formats (including print, electronic, and online), in sublicensed and reprinted versions (including translations and derived works), and in other works and products under open access license. To respect patients' and any other individual's privacy, please do not send signed forms. The journal reserves the right to ask authors to provide signed forms if necessary.

Ethical Guidelines for the Use of Animals in Research

The editors will require that the benefits potentially derived from any research causing harm to animals are significant in relation to any cost endured by animals, and that procedures followed are unlikely to

cause offense to the majority of readers. Authors should particularly ensure that their research complies with the commonly-accepted '3Rs':

- Replacement of animals by alternatives wherever possible,
- Reduction in number of animals used, and
- Refinement of experimental conditions and procedures to minimize the harm to animals.

Any experimental work must also have been conducted in accordance with relevant national legislation on the use of animals for research. For further guidance authors should refer to the Code of Practice for the Housing and Care of Animals Used in Scientific Procedures [1].

Manuscripts containing original descriptions of research conducted in experimental animals must contain details of approval by a properly constituted research ethics committee. As a minimum, the project identification code, date of approval and name of the ethics committee or institutional review board should be stated in Section 'Institutional Review Board Statement'.

JCM endorses the ARRIVE guidelines (www.nc3rs.org.uk/ARRIVE) for reporting experiments using live animals. Authors and reviewers can use the ARRIVE guidelines as a checklist, which can be found at www.nc3rs.org.uk/ARRIVEchecklist.

1. Home Office. Animals (Scientific Procedures) Act 1986. Code of Practice for the Housing and Care of Animals Used in Scientific Procedures. Available online:

<http://www.official-documents.gov.uk/document/hc8889/hc01/0107/0107.pdf>.

Research Involving Cell Lines

Methods sections for submissions reporting on research with cell lines should state the origin of any cell lines. For established cell lines the provenance should be stated and references must also be given to either a published paper or to a commercial source. If previously unpublished de novo cell lines were used, including those gifted from another laboratory, details of institutional review board or ethics committee approval must be given, and confirmation of written informed consent must be provided if the line is of human origin.

An example of Ethical Statements:

The HCT116 cell line was obtained from XXXX. The MLH1+ cell line was provided by XXXXX, Ltd. The DLD-1 cell line was obtained from Dr. XXXX. The DR-GFP and SA-GFP reporter plasmids were obtained from Dr. XXX and the Rad51K133A expression vector was obtained from Dr. XXXX.

Clinical Trials Registration

Registration

MDPI follows the International Committee of Medical Journal Editors (ICMJE) guidelines which require and recommend registration of clinical trials in a public trials registry at or before the time of first patient enrollment as a condition of consideration for publication.

Purely observational studies do not require registration. A clinical trial not only refers to studies that take place in a hospital or involve pharmaceuticals, but also refer to all studies which involve participant randomization and group classification in the context of the intervention under assessment.

Authors are strongly encouraged to pre-register clinical trials with an international clinical trials register and cite a reference to the registration in the abstract and Methods section. Suitable databases include

clinicaltrials.gov, the EU Clinical Trials Register and those listed by the World Health Organisation International Clinical Trials Registry Platform.

Approval to conduct a study from an independent local, regional, or national review body is not equivalent to prospective clinical trial registration. MDPI reserves the right to decline any paper without trial registration for further peer-review. However, if the study protocol has been published before the enrolment, the registration can be waived with correct citation of the published protocol.

CONSORT Statement

MDPI requires a completed CONSORT 2010 checklist and flow diagram as a condition of submission when reporting the results of a randomized trial. Templates for these can be found here or on the CONSORT website (<http://www.consort-statement.org>) which also describes several CONSORT checklist extensions for different designs and types of data beyond two group parallel trials. At minimum, your article should report the content addressed by each item of the checklist.

Borders and Territories

Potential disputes over borders and territories may have particular relevance for authors in describing their research or in an author or editor correspondence address, and should be respected. Content decisions are an editorial matter and where there is a potential or perceived dispute or complaint, the editorial team will attempt to find a resolution that satisfies parties involved.

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Publication Ethics Statement

JCM is a member of the Committee on Publication Ethics ([COPE](#)). We fully adhere to its [Code of Conduct](#) and to its [Best Practice Guidelines](#).

The editors of this journal enforce a rigorous peer-review process together with strict ethical policies and standards to ensure to add high quality scientific works to the field of scholarly publication.

Unfortunately, cases of plagiarism, data falsification, image manipulation, inappropriate authorship credit, and the like, do arise. The editors of JCM take such publishing ethics issues very seriously and are trained to proceed in such cases with a zero tolerance policy.

Authors wishing to publish their papers in JCM must abide to the following:

- Any facts that might be perceived as a possible conflict of interest of the author(s) must be disclosed in the paper prior to submission.
- Authors should accurately present their research findings and include an objective discussion of the significance of their findings.
- Data and methods used in the research need to be presented in sufficient detail in the paper, so that other researchers can replicate the work.
- Raw data should preferably be publicly deposited by the authors before submission of their manuscript. Authors need to at least have the raw data readily available for presentation to the referees and the editors of the journal, if requested. Authors need to ensure appropriate measures are taken so that raw data is retained in full for a reasonable time after publication.
- Simultaneous submission of manuscripts to more than one journal is not tolerated.
- Republishing content that is not novel is not tolerated (for example, an English translation of a paper that is already published in another language will not be accepted).

- If errors and inaccuracies are found by the authors after publication of their paper, they need to be promptly communicated to the editors of this journal so that appropriate actions can be taken. Please refer to our policy regarding Updating Published Papers.

- Your manuscript should not contain any information that has already been published. If you include already published figures or images, please obtain the necessary permission from the copyright holder to publish under the CC-BY license. For further information, see the Rights and Permissions page.

- Plagiarism, data fabrication and image manipulation are not tolerated.

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Plagiarism includes copying text, ideas, images, or data from another source, even from your own publications, without giving any credit to the original source.

Reuse of text that is copied from another source must be between quotes and the original source must be cited. If a study's design or the manuscript's structure or language has been inspired by previous works, these works must be explicitly cited.

If plagiarism is detected during the peer review process, the manuscript may be rejected. If plagiarism is detected after publication, we may publish a correction or retract the paper.

- o **Image files must not be manipulated or adjusted in any way** that could lead to misinterpretation of the information provided by the original image.

Irregular manipulation includes: 1) introduction, enhancement, moving, or removing features from the original image; 2) grouping of images that should obviously be presented separately (e.g., from different parts of the same gel, or from different gels); or 3) modifying the contrast, brightness or color balance to obscure, eliminate or enhance some information.

If irregular image manipulation is identified and confirmed during the peer review process, we may reject the manuscript. If irregular image manipulation is identified and confirmed after publication, we may correct or retract the paper.

Our in-house editors will investigate any allegations of publication misconduct and may contact the authors' institutions or funders if necessary. If evidence of misconduct is found, appropriate action will be taken to correct or retract the publication. Authors are expected to comply with the best ethical publication practices when publishing with MDPI.

Citation Policy

Authors should ensure that where material is taken from other sources (including their own published writing) the source is clearly cited and that where appropriate permission is obtained.

Authors should not engage in excessive self-citation of their own work.

Authors should not copy references from other publications if they have not read the cited work.

Authors should not preferentially cite their own or their friends', peers', or institution's publications.

Authors should not cite advertisements or advertorial material.

In accordance with COPE guidelines, we expect that "original wording taken directly from publications by other researchers should appear in quotation marks with the appropriate citations." This condition also applies to an author's own work. COPE have produced a discussion document on citation manipulation with recommendations for best practice.

Reviewer Suggestions

During the submission process, please suggest three potential reviewers with the appropriate expertise to review the manuscript. The editors will not necessarily approach these referees. Please provide detailed contact information (address, homepage, phone, e-mail address). The proposed referees should neither be current collaborators of the co-authors nor have published with any of the co-authors of the manuscript within the last five years. Proposed reviewers should be from different institutions to the authors. You may identify appropriate Editorial Board members of the journal as potential reviewers. You may suggest reviewers from among the authors that you frequently cite in your paper.

English Corrections

To facilitate proper peer-reviewing of your manuscript, it is essential that it is submitted in grammatically correct English. Advice on some specific language points can be found [here](#).

If you are not a native English speaker, we recommend that you have your manuscript professionally edited before submission or read by a native English-speaking colleague. This can be carried out by MDPI's English editing service. Professional editing will enable reviewers and future readers to more easily read and assess the content of submitted manuscripts. All accepted manuscripts undergo language editing, however an additional fee will be charged to authors if very extensive English corrections must be made by the Editorial Office: pricing is according to the service [here](#).

Preprints and Conference Papers

JCM accepts submissions that have previously been made available as preprints provided that they have not undergone peer review. A preprint is a draft version of a paper made available online before submission to a journal.

MDPI operates Preprints, a preprint server to which submitted papers can be uploaded directly after completing journal submission. Note that Preprints operates independently of the journal and posting a preprint does not affect the peer review process. Check the Preprints instructions for authors for further information.

Expanded and high-quality conference papers can be considered as articles if they fulfill the following requirements: (1) the paper should be expanded to the size of a research article; (2) the conference paper should be cited and noted on the first page of the paper; (3) if the authors do not hold the copyright of the published conference paper, authors should seek the appropriate permission from the copyright holder; (4) authors are asked to disclose that it is conference paper in their cover letter and include a statement on what has been changed compared to the original conference paper. JCM does not publish pilot studies or studies with inadequate statistical power.

Unpublished conference papers that do not meet the above conditions are recommended to be submitted to the Proceedings series of journals:

Proceedings: <https://www.mdpi.com/journal/proceedings>

Authorship

MDPI follows the International Committee of Medical Journal Editors (ICMJE) guidelines which state that, in order to qualify for authorship of a manuscript, the following criteria should be observed:

- Substantial contributions to the conception or design of the work; or the acquisition, analysis, or interpretation of data for the work; AND

- Drafting the work or revising it critically for important intellectual content; AND
- Final approval of the version to be published; AND
- Agreement to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Those who contributed to the work but do not qualify for authorship should be listed in the acknowledgments. More detailed guidance on authorship is given by the International Council of Medical Journal Editors (ICMJE).

Any change to the author list should be approved by all authors including any who have been removed from the list. The corresponding author should act as a point of contact between the editor and the other authors and should keep co-authors informed and involve them in major decisions about the publication. We reserve the right to request confirmation that all authors meet the authorship conditions.

Reviewers Recommendation

Authors can recommend potential reviewers. Journal editors will check to make sure there are no conflicts of interest before contacting those reviewers, and will not consider those with competing interests. Reviewers are asked to declare any conflicts of interest. Authors can also enter the names of potential peer reviewers they wish to exclude from consideration in the peer review of their manuscript, during the initial submission progress. The editorial team will respect these requests so long as this does not interfere with the objective and thorough assessment of the submission.

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Lack of Interference With Editorial Decisions

Editorial independence is of utmost importance and MDPI does not interfere with editorial decisions. All articles published by MDPI are peer reviewed and assessed by our independent editorial boards, and MDPI staff are not involved in decisions to accept manuscripts. When making an editorial decision, we expect the academic editor to make their decision based only upon:

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- Adequacy of reviewer comments and author response;
- Overall scientific quality of the paper.

In all of our journals, in every aspect of operation, MDPI policies are informed by the mission to make science and research findings open and accessible as widely and rapidly as possible.

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Editorial staff or editors shall not be involved in processing their own academic work. Submissions authored by editorial staff/editors will be assigned to at least two independent outside reviewers. Decisions will be made by other Editorial Board Members who do not have a conflict of interest with the author. Journal staff are not involved in the processing of their own work submitted to any MDPI journals.

Conflict of Interests

According to The International Committee of Medical Journal Editors, “Authors should avoid entering into agreements with study sponsors, both for-profit and non-profit, that interfere with authors’ access to all of the study’s data or that interfere with their ability to analyze and interpret the data and to prepare and publish manuscripts independently when and where they choose.”

All authors must disclose all relationships or interests that could inappropriately influence or bias their work. Examples of potential conflicts of interest include but are not limited to financial interests (such as membership, employment, consultancies, stocks/shares ownership, honoraria, grants or other funding, paid expert testimonies and patent-licensing arrangements) and non-financial interests (such as personal or professional relationships, affiliations, personal beliefs).

Authors can disclose potential conflicts of interest via the online submission system during the submission process. Declarations regarding conflicts of interest can also be collected via the MDPI disclosure form. The corresponding author must include a summary statement in the manuscript in a separate section “Conflicts of Interest” placed just before the reference list. The statement should reflect all the collected potential conflict of interest disclosures in the form.

See below for examples of disclosures:

Conflicts of Interest: Author A has received research grants from Company A. Author B has received a speaker honorarium from Company X and owns stocks in Company Y. Author C has been involved as a consultant and expert witness in Company Z. Author D is the inventor of patent X.

If no conflicts exist, the authors should state:

Conflicts of Interest: The authors declare no conflicts of interest.

Editorial Procedures and Peer-Review

Initial Checks

All submitted manuscripts received by the Editorial Office will be checked by a professional in-house Managing Editor to determine whether they are properly prepared and whether they follow the ethical policies of the journal, including those for human and animal experimentation. Manuscripts that do not fit the journal's ethics policy or do not meet the standards of the journal will be rejected before peer-review. Manuscripts that are not properly prepared will be returned to the authors for revision and resubmission. After these checks, the Managing Editor will consult the journals’ Editor-in-Chief or Associate Editors to determine whether the manuscript fits the scope of the journal and whether it is scientifically sound. No judgment on the potential impact of the work will be made at this stage. Reject decisions at this stage will be verified by the Editor-in-Chief.

Peer-Review

Once a manuscript passes the initial checks, it will be assigned to at least two independent experts for peer-review. A single-blind review is applied, where authors' identities are known to reviewers. Peer review comments are confidential and will only be disclosed with the express agreement of the reviewer.

In the case of regular submissions, in-house assistant editors will invite experts, including recommendations by an academic editor. These experts may also include Editorial Board Members and Guest Editors of the journal. Potential reviewers suggested by the authors may also be considered. Reviewers should not have published with any of the co-authors during the past five years and should not currently work or collaborate with any of the institutions of the co-authors of the submitted manuscript.

Optional Open Peer-Review

The journal operates optional open peer-review: Authors are given the option for all review reports and editorial decisions to be published alongside their manuscript. In addition, reviewers can sign their review, i.e., identify themselves in the published review reports. Authors can alter their choice for open review at any time before publication, but once the paper has been published changes will only be made at the discretion of the Publisher and Editor-in-Chief. We encourage authors to take advantage of this opportunity as proof of the rigorous process employed in publishing their research. To guarantee impartial refereeing, the names of referees will be revealed only if the referees agree to do so, and after a paper has been accepted for publication.

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All the articles, reviews and communications published in MDPI journals go through the peer-review process and receive at least two reviews. The in-house editor will communicate the decision of the academic editor, which will be one of the following:

- **Accept after Minor Revisions:** The paper is in principle accepted after revision based on the reviewer's comments. Authors are given five days for minor revisions.
- **Reconsider after Major Revisions:** The acceptance of the manuscript would depend on the revisions. The author needs to provide a point by point response or provide a rebuttal if some of the reviewer's comments cannot be revised. Usually, only one round of major revisions is allowed. Authors will be asked to resubmit the revised paper within a suitable time frame, and the revised version will be returned to the reviewer for further comments.
- **Reject and Encourage Resubmission:** If additional experiments are needed to support the conclusions, the manuscript will be rejected and the authors will be encouraged to re-submit the paper once further experiments have been conducted.
- **Reject:** The article has serious flaws, and/or makes no original significant contribution. No offer of resubmission to the journal is provided.

All reviewer comments should be responded to in a point-by-point fashion. Where the authors disagree with a reviewer, they must provide a clear response.

Author Appeals

Authors may appeal a rejection by sending an e-mail to the Editorial Office of the journal. The appeal must provide a detailed justification, including point-by-point responses to the reviewers' and/or Editor's comments. The Managing Editor of the journal will forward the manuscript and related information (including the identities of the referees) to the Editor-in-Chief, Associate Editor, or Editorial Board member. The academic Editor being consulted will be asked to give an advisory recommendation on the manuscript and may recommend acceptance, further peer-review, or uphold the original rejection decision. A reject decision at this stage is final and cannot be reversed.

In the case of a special issue, the Managing Editor of the journal will forward the manuscript and related information (including the identities of the referees) to the Editor-in-Chief who will be asked to give an advisory recommendation on the manuscript and may recommend acceptance, further peer-review, or uphold the original rejection decision. A reject decision at this stage will be final and cannot be reversed.

Production and Publication

Once accepted, the manuscript will undergo professional copy-editing, English editing, proofreading by the authors, final corrections, pagination, and, publication on the www.mdpi.com website.

Promoting Equity, Diversity and Inclusiveness Within MDPI Journals

Our Managing Editors encourage the Editors-in-Chief and Associate Editors to appoint diverse expert Editorial Boards. This is also reflective in our multi-national and inclusive workplace. We are proud to create equal opportunities without regard to gender, ethnicity, sexual orientation, age, religion, or socio-economic status. There is no place for discrimination in our workplace and editors of MDPI journals are to uphold these principles in high regard.

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