Medicinski podmladak

Medical Youth

Mini review article

ACCUMULATION OF METAL(LOID)S IN MYOCARDIAL TISSUE AND THE MECHANISMS UNDERLYING THEIR CARDIOTOXIC EFFECTS

TALOŽENJE METALA (I METALOIDA) U SRČANOM TKIVU I MEHANIZMI KOJI LEŽE U OSNOVI NJIHOVIH KARDIOTOKSIČNIH EFEKATA

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Abstract

Heavy metals could exert a strong cardiotoxic effect, since cardiomyocytes are vulnerable cells in general, very sensitive to heavy metals-induced toxicity. The correlation between exposure to heavy metals and their contribution to the pathophysiology of diverse cardiovascular disorders, such as coronary artery disease (CAD) and cardiomyopathies (CMPs), has gained recognition primarily through clinical investigations wherein metal(loid) levels were quantified in the blood or urine of individuals afflicted with aforementioned disorders. However, a crucial perspective is absent due to lack of studies that investigate the accumulation of heavy metals within cardiac tissue. These studies, whether post-mortem or involving heart samples obtained during invasive procedures, are currently lacking. To achieve a comprehensive understanding of the potential involvement of metal(loid)s in the genesis of e.g. CMPs or CAD, these inquiries are indispensable. Furthermore, certain comorbidities like iron deficiency may expedite the bioaccumulation of myocardial heavy metals by augmenting the density of transferrin receptor 1 (TfR1). The impact of heavy metals on the heart's contractile machinery, coupled with their potential to initiate mitochondrial apoptosis through triggered pathways, forms part of the intricate pathophysiological landscape. Central to these mechanisms is the generation of reactive oxygen species (ROS) and the peroxidation of macromolecules.

This review highlights the research findings on the bioaccumulation of heavy metals within the myocardium and elucidates the molecular mechanisms through which metal(loid)s induce cardiotoxicity.

Keywords:
metals bioaccumulation,
myocardium,
iron deficiency

doi:10.5937/mp74-46164
Editorial board: podmladak.med.bg@gmail.com
e-ISSN: 2466-5525

Sažetak

Teški metali imaju snažan kardiotoksični efekat, a kardiomiociti su, uopšteno govoreći, osetljive ćelije, te su stoga veoma podložne toksičnosti uzrokovanoj prisustvom teških metala. Veza između izloženosti teškim metalima i njihovog doprinosa u patofiziologiji različitih kardiovaskularnih poremećaja, kao što su koronarna bolest srca (engl. *coronary artery disease* - CAD) i kardiomiopatije (engl. *cardiomyophaties* - CMP), prepoznata je pretežno putem kliničkih istraživanja u kojima su nivoi metala ili metaloida mereni u krvi ili urinu osoba koje su imale pomenuta oboljenja. Ključnu kariku, koja bi doprinela boljem razumevanju povezanosti teških metala i nastanku kardioloških oboljenja, predstavljaju studije koje ispituju taloženje teških metala unutar srčanog tkiva. Ovakve studije, bilo da su analizirani uzorci dobijeni postmortem ili tokom invazivnih procedura u pacijenata, trenutno nedostaju. Ovakva istraživanja su neophodna kako bi se postiglo sveobuhvatno razumevanje potencijalne uloge metala i metaloida u nastanku CMP ili CAD. Osim toga, određeni komorbiditeti, kao što je nedostatak gvožđa, mogu ubrzati taloženje teških metala u srčanom mišiću povećavanjem gustine receptora za transferin (engl. *transferrin receptor 1* - TfR1). Uticaj teških metala na kontraktilni aparat srca, zajedno sa njihovim potencijalom da započnu mitohondrijalnu apoptozu, čini deo složenog patofiziološkog pejzaža. Bazični mehanizmi uključeni u oštećenja usled prisustva teških metala su generisanje reaktivnih vrsta kiseonika (engl. *reactive oxygen species* - ROS) i peroksidacija makromolekula.

U ovom članku ćemo se osvrnuti na studije koje su analizirale taloženje teških metala unutar miokarda, kao i na studije koje su se bavile molekularnim mehanizmima putem kojih metali ili metaloidi izazivaju kardiotoksičnost.

Ključne reči:
taloženje metala,
miokard,
nedostatak gvožđa

Introduction

Optimal heart function is imperative for the effective operation of the entire body and the oxygenation of peripheral tissues. The left ventricle (LV) is responsible for propelling blood into the systemic circulation, and any weakening of its functionality could precipitate widespread bodily distress. While it is widely acknowledged that prolonged and untreated high blood pressure can trigger LV issues, due to the gravity of the matter, all factors influencing LV function should be thoroughly assessed, including environmental factors such as heavy metals.

Humans encounter heavy metals in the course of their daily activities; tap water is often tainted with arsenic (1), aluminum is an integral component of various cosmetics, including antiperspirants (2), and rice in Asia as well as vegetables in Europe are frequently found to be contaminated with cadmium (3,4). Presently, it is established that elevated blood levels of cadmium may be linked to coronary artery atherosclerosis (5), whereas individuals with coronary artery disease exhibit heightened levels of mercury, lead, and cadmium in their bloodstream (6). Fetal aluminum exposure has been associated with the appearance of congenital heart disease (7). On the side, individuals with dilated cardiomyopathy manifest escalated urine concentrations of chromium, manganese, cobalt, copper, zinc, selenium, cadmium, tin, antimony, thallium, and uranium compared to control subjects. The association between exposure to heavy metals, their bioavailability, and their accumulation in myocardial tissue warrants further exploration. Consequently, the objective of this review is to provide a concise overview of certain facets of the bioaccumulation of metal(loid)s in the myocardium, and to deliberate upon potential factors that may facilitate the uptake of heavy metals by the myocardium.

Bioaccumulation of metal(loid)s in myocardium: an anatomical mapping

Recognizing that the detrimental impacts induced by the bioaccumulation of heavy metals exhibit a direct correlation with the concentrations of these metals in tissues (8), two fundamental questions come to the fore. The first inquiry revolves around whether metal(loid)s, particularly heavy metals, accumulate uniformly throughout the entirety of the myocardium, or if specific regions display a heightened susceptibility to the uptake of heavy metals. The second query emerges from the pandemic proportions of heart disorders, such as coronary artery disease and heart failure, and the yet undiscovered role of heavy metals in the pathogenesis of these conditions. As a result, a comprehensive investigation becomes imperative.

Employing laser ablation inductively coupled plasma mass spectrometry (LA-ICPMS), Egger and collaborators conducted an analysis of cadmium concentrations in 40 distinct human tissues, including the left ventricle, sourced from four body donors at the Institute of Anatomy (9). Their findings revealed that the mean cadmium concentration among these four individuals was 35 µg/kg, with a range spanning from 21.8 to 72.6 µg/kg. However, they observed that, considering all 40 tissue types, cadmium levels exhibited variances of approximately 20,000-fold between different tissues. While the liver displays some of the highest levels of cadmium, hepatocytes possess a notable regenerative capacity and do not operate continuously

throughout an entire lifetime. Conversely, cardiomyocytes' regenerative potential is considerably limited. Moreover, cardiomyocytes are about 100 times more sensitive to cadmium-induced toxicity in comparison to hepatocytes (10). This underscores the necessity to explore whether metals accumulate uniformly within the cardiac muscle.

Similarly, Cirovic et al. undertook a study to measure the concentrations of magnesium (Mg), calcium (Ca), cadmium (Cd), lead (Pb), iron (Fe), zinc (Zn), copper (Cu), manganese (Mn), nickel (Ni), mercury (Hg), and arsenic (As) in two distinct areas of the left ventricle myocardium. The heart samples were procured from the Institute of Anatomy. The researchers discovered elevated concentrations of lead and arsenic at the apex of the left ventricle in comparison to the base of the left ventricle, suggesting that specific metal(loid)s tend to accumulate to a greater extent in particular regions of the myocardium (11). Utilizing samples from individuals without heart disorders, they employed LA-ICPMS for their investigation. Additionally, Becker et al. conducted a study wherein anatomical mapping, specifically bioimaging of metals distribution, was performed on healthy male mice ventricles using the same technique (12). Their research unveiled a non-uniform distribution of all assessed elements (zinc, copper, manganese, magnesium, iron, etc.) throughout the myocardium of the ventricles. Thus, both of the aforementioned studies highlighted the potential existence of a distinct pattern of metal distribution within the myocardium.

Conversely, an intriguing study by Frustaci et al. focused on the measurement of silver (Ag), arsenic (As), gold (Au), barium (Ba), calcium (Ca), cadmium (Cd), cerium (Ce), cobalt (Co), chromium (Cr), cesium (Cs), europium (Eu), iron (Fe), hafnium (Hf), mercury (Hg), iridium (Ir), lanthanum (La), lutetium (Lu), molybdenum (Mo), nickel (Ni), rubidium (Rb), antimony (Sb), scandium (Sc), selenium (Se), samarium (Sm), tin (Sn), strontium (Sr), tantalum (Ta), terbium (Tb), thorium (Th), uranium (U), ytterbium (Yb), zinc (Zn), and zirconium (Zr) concentrations in 13 individuals (9 males, 4 females, mean age 50.9 ± 10.1 years) afflicted with idiopathic dilated cardiomyopathy. The method employed for analysis was neutron activation analysis, and endomyocardial biopsies were procured from the left ventricular apex during cardiac catheterization. Their findings illuminated values several thousand times higher for antimony and mercury in the left ventricular apex of subjects suffering from idiopathic dilated cardiomyopathy, as compared to the control group (13).

Influential factors affecting the rate of metal bioaccumulation in myocardial tissue

The ingress of heavy metals into the body occurs through both the gastrointestinal and respiratory routes. However, due to inevitable food contamination by various metal(loid)s (such as chromium, nickel, copper, zinc, arsenic, cadmium, and lead), the gastrointestinal tract emerges as the foremost conduit for heavy metal absorption, particularly in instances of iron deficiency. Notably, iron deficiency in both adults and children has been shown to result in elevated blood levels of diverse divalent cations such as manganese (14), lead (15) and cadmium (16). This heightened load of heavy metals in the human body is attributed to the overexpression of divalent metal transporter 1 (DMT1), initially an iron carrier, yet capable of up taking a broad array of metals (including Fe, Zn, Mn, Co, Cd, Cu, Ni, and Pb) from ingested food, due to its presence in the small intestine, particularly the duodenum (17).

Upon absorption from the intestinal lumen, roughly 50% of cadmium is conveyed through the bloodstream, binding with transferrin, while the remainder predominantly binds to albumin (18). Consequently, it is anticipated that tissues with a higher density of transferrin receptor 1 (TfR1) will accumulate heavy metals to a greater degree compared to tissues with lower TfR1 density. Intriguingly, Cabrera et al. scrutinized the expression levels of genes related to iron metabolism in 69 individuals (among whom 28% had iron deficiency) undergoing elective coronary bypass surgery (CABG). During these procedures, core needle biopsies were obtained from both the left and right ventricles. They discerned an augmented expression of both DMT1 and TfR1 in the myocardium of individuals with iron deficiency, particularly those with absolute iron deficiency (19). Based on this elucidated mechanism, it is evident that under conditions of iron deficiency, the myocardium accumulates heavy metals to a greater extent (**figure 1**). Importantly, it must be reiterated that cardiomyocytes are exceptionally sensitive to the detrimental effects caused by heavy metals. Consequently, in the context of prolonged iron deficiency, while the body absorbs heavy metals from ingested food, the heart undergoes an exacerbated intake of heavy metals. The potential role of cadmium and other metals in the pathogenesis of heart disorders in individuals with iron deficiency has been briefly explored by Cirovic et al. (20) and Savarese et al. (21).

Metal(loid) induced-cardiotoxicity

At times, detrimental effects attributed to metals become apparent during clinical assessments. Specifically, it is recognized that individuals afflicted with iron overload disorders, such as hemochromatosis, develop cardiac hemochromatosis (dilated cardiomyopathy) due to iron toxicity. Once all ferritin molecules within the cell become saturated, free Fe^{2+} inflicts damage upon cardiomyocytes through a process known as Fenton's reaction.

Knowledge on the cellular mechanisms underlying heavy metal-induced cardiotoxicity is primarily documented through in vivo experiments (22, 23) and in vitro studies (10, 24). Gerzan et al. exposed male rats to lead acetate three times a week over a span of 35 days, demonstrating that lead impaired the mechanical characteristics of myosin in both the atria and ventricles (23). Also, in another in vivo study, Furieri et al. demonstrated that exposure to $HgCl_2$ can induce alteration of myocardial

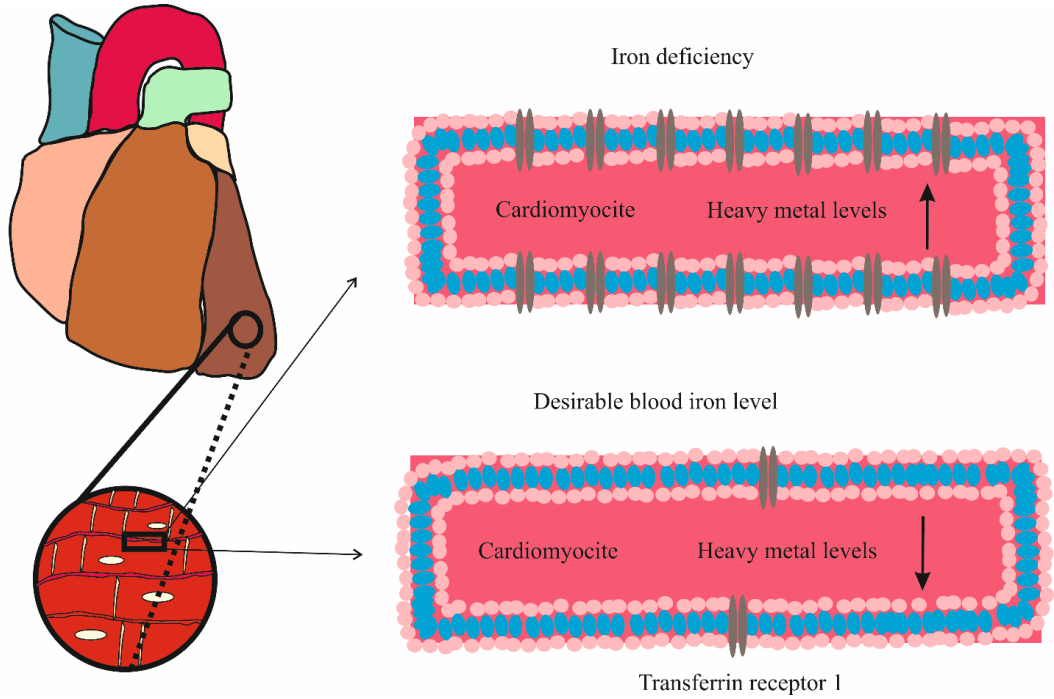

Figure 1. Schematic representation of mechanisms involved in heavy metals myocardial bioaccumulation in the case of iron deficiency.

contractile apparatus (25). In a separate study, Zhao et al. conducted an experiment involving 10 swine, categorized into two groups: control and cadmium-fed ($CdCl_2$, 20 mg Cd/kg diet) (26). Cadmium concentrations were measured post-experiment, revealing that cadmium-fed swine exhibited concentrations over 8 times higher. Moreover, they found that cadmium triggered the mitochondrial apoptosis pathway within the swine myocardium. The molecular mechanism driving apoptosis involved an elevation in CYP450s expression, ultimately leading to the generation of reactive oxygen species (ROS). Intriguingly, copper, an integral component of superoxide dismutase capable of triggering myocardial regeneration, has the potential to induce myocardial damage as well (27, 28). Pan et al. conducted an experiment in which authors included 32 male mice divided to two groups, controls and copper-treated group. Authors performed a series of biochemical analyses, echocardiographic measurements, histological analyses etc. Their research reported several key findings: i) heart rate (HR), left ventricular ejection fraction (LVEF), left ventricular fractional shortening (LVFS) are found to be lower in copper-treated mice; ii) authors described several histological alterations such as significantly swollen mitochondria; and iii) finally, the expression of BAX gene (pro-apoptotic member of the Bcl-2 gene family) cleaved caspase-3 were upregulated (28). The detrimental effects of heavy metals on cardiomyocytes are manifested through an increase in creatine kinase (CK) and lactate dehydrogenase (LDH) levels in the blood of animals exposed to arsenic trioxide (29). Elevated myocardial malondialdehyde activity (MDA) has been identified as a mechanism involved in heavy metal-induced cardiotoxicity in numerous in vivo studies (30, 31). Myocardial malondialdehyde activity serves as a marker of oxidative stress, and the oxidative stress induced by the generation of free radicals represents a fundamental mechanism of heavy metal-induced cardiotoxicity. Shen et al. cultivated H9 human embryonic stem cells until they fully differentiated into cardiomyocytes. Subsequently, the authors exposed these cells to a wide range of $CdCl_2$ doses, ranging from 0.1 to 100 μmol/L, for 24 hours. They demonstrated a significant reduction in cardiomyocytes viability and an increase in ROS generation in $CdCl_2$-treated H9-CMs, following a dose-dependent pattern (24). Das et al. administered cadmium orally to 8-week-old male Sprague-Dawley rats at a dosage of 15 mg/kg of body weight for a duration of 10 weeks. Their findings revealed that animals treated with cadmium for 10 weeks exhibited notably upregulated levels of pro-inflammatory mediators: interleukin-6 (IL-6), interleukin-1β (IL-1β), nuclear factor-κB (NF-κB), and tumor necrosis factor-alpha (TNF-α) (32). Adit et al. demonstrated that oral exposure to sodium arsenite for 28 days may induce several alterations in the myocardium of Sprague-Dawley rats. Initially, they observed that sodium arsenite-induced treatment led to an increase in relative heart weight. Additionally, the heart rate was lower in arsenite-treated animals, accompanied by significant alterations in electrocardiographic parameters (such as increased intervals QRS, QT, QTc, and RR). Exposure to sodium arsenite also altered the lipid profile, manifesting as increments in the following parameters: total cholesterol, triglyceride, LDL, and VLDL levels, whereas HDL was significantly decreased (33). This indicates that arsenic exposure could promote atherosclerosis. Furthermore, treatment via oral administration of 5 mg/kg, the dosage used by the authors in this study, downregulated concentrations of antioxidants such as superoxide dismutase and glutathione. This downregulation promoted the accumulation of oxidative stress markers (MDA and NO) (33).

Conclusion

Metal(loid)s exhibit an uneven distribution in the myocardium. A comprehensive mapping of the specific regions within the myocardium that are disproportionately impacted by metal(loid)s bioaccumulation remains incomplete. A more nuanced comprehension of this matter could not only enhance our understanding of heavy metal metabolism but also illuminate the potential role of metal(loid)s in the pathophysiology of diverse cardiac disorders.

The hypothesis posits that iron deficiency might contribute to the emergence of various cardiac disorders by elevating the body's burden of heavy metals. The influence of heavy metals on the heart's contractile machinery and their potential to activate the mitochondrial apoptosis pathway constitute key aspects. Nonetheless, fundamental pathophysiological mechanisms implicate the generation of reactive oxygen species (ROS) and the peroxidation of macromolecules.

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