The Big 4 Toxic Metals & Their Impact on Patient Health

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August 15, 2016

What are the Big 4 Heavy Metals?

- ATSDR (Agency for Toxic Substances and Disease Registry) priority list of hazardous Substances
  - #1 Arsenic
  - #2 Lead
  - #3 Mercury
  - #7 Cadmium

Sources:
1) http://www.atsdr.cdc.gov/spl

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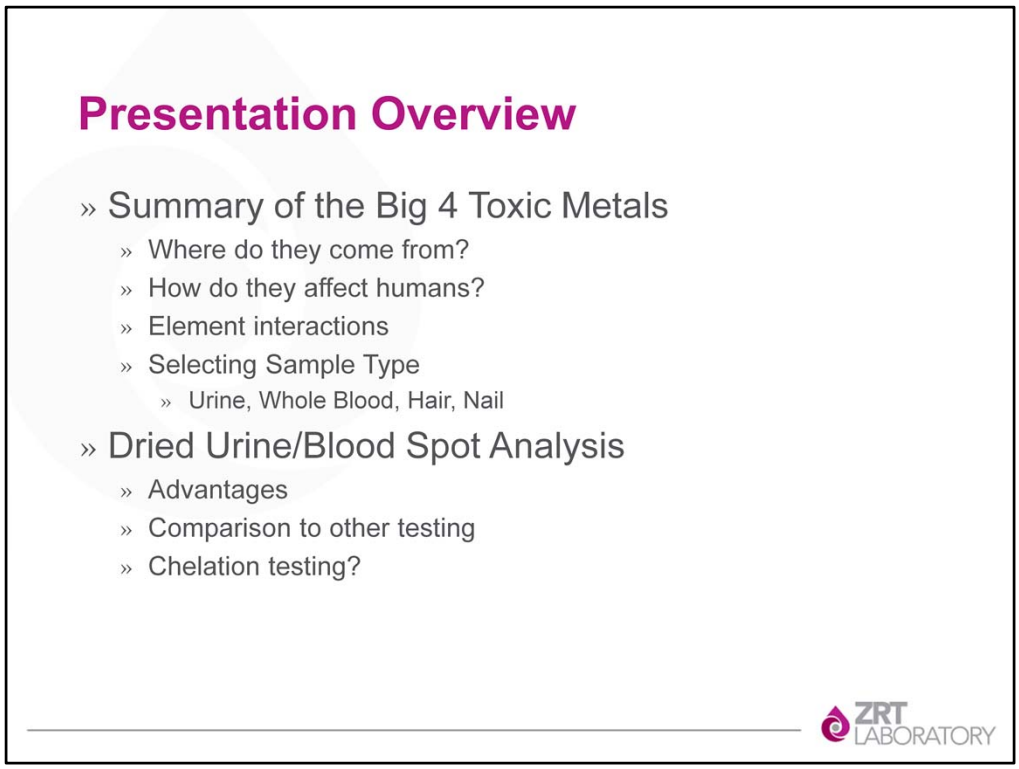
What at the big 4 heavy metals?

The agency for toxic substances and disease registry publishes a list every two years that prioritizes substances based on their frequency, toxicity, and potential for human exposure.

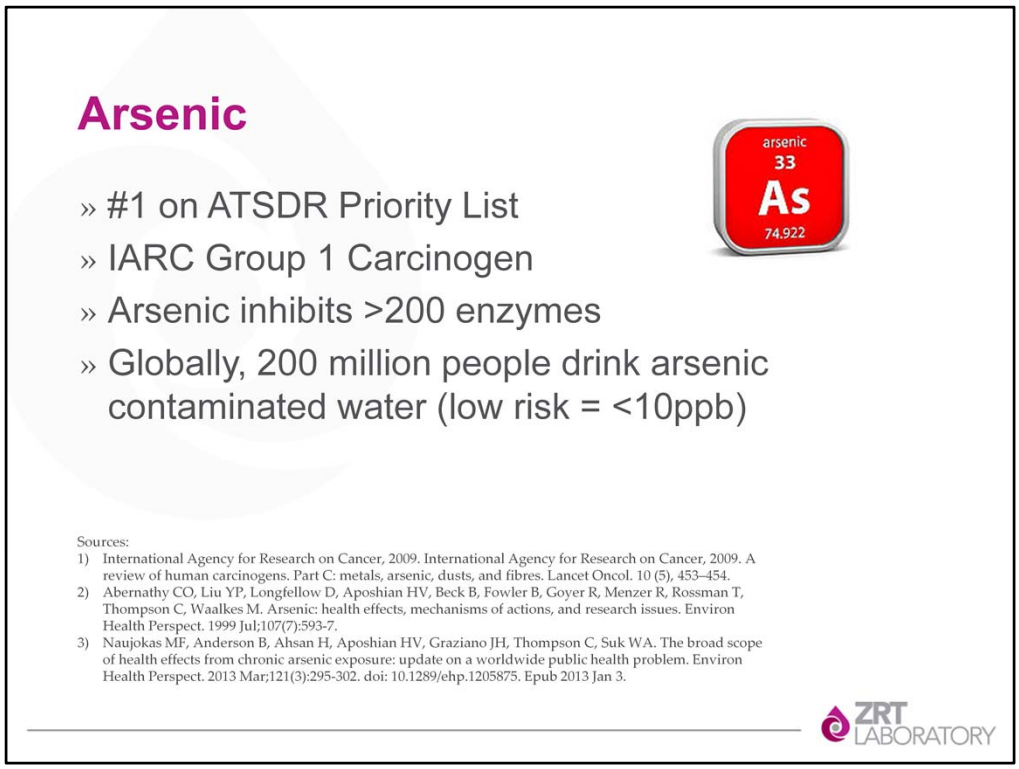
The most recent list was formulated in 2015 . Out of 275 hazardous substances, 4 heavy metals ranked in the top 10. These are Arsenic at #1, Lead at #2, Mercury at #3, and Cadmium at #7.

These are the big 4 heavy metals.



This presentation will cover what you need to know about the big 4 toxic metals. It will include summaries of the individual elements, how essential elements interact, the differences between urine, whole blood, hair and nail sample types, along with a discussion about reference ranges are and how they can affect results.

We will end the presentation talking about our dried urine and blood spot element analysis, which is unique to ZRT Laboratory, and the benefits of this type of testing for toxic and essential elements.



We will begin with Arsenic

Arsenic is #1 on the A T S D R hazardous substances priority list for very good reasons..

It is the only group 1 carcinogen established by the International Agency for Research on Cancer that is active by both ingestion and inhalation.

Extensive research has shown that arsenic can inhibit over 200 enzymes in the body.

The Environmental Protection Agency and World Health organization set a recommended limit of 10ug of arsenic per liter of water, yet over 200 million people globally are exposed to high levels of arsenic from home wells and public water sources.

In the US, several states are know to have high arsenic in aquafers. Those include Arizona, California and Nevada, but high arsenic in wells is not limited to these states.

Arsenic can be broken down into three separate species. Inorganic, Methylated, and Organic. The toxicity of each of these species varies significantly, along with the source.

There are a large number of inorganic, methylated, and organic arsenic species, many of which have not been identified or studied. This presentation will focus on those most commonly encountered.

The most common inorganic species are Trivalent Arsenic and Pentavalent Arsenic.

The most common methylated species are Monomethyl Arsenic and Dimethyl Arsenic.

The most common organic species are Arsenobetaine and Arsenocholine.

Inorganic arsenic species are far more dangerous than organic arsenic species. The primary damaging action of inorganic arsenic is through oxidative stress and DNA damage.

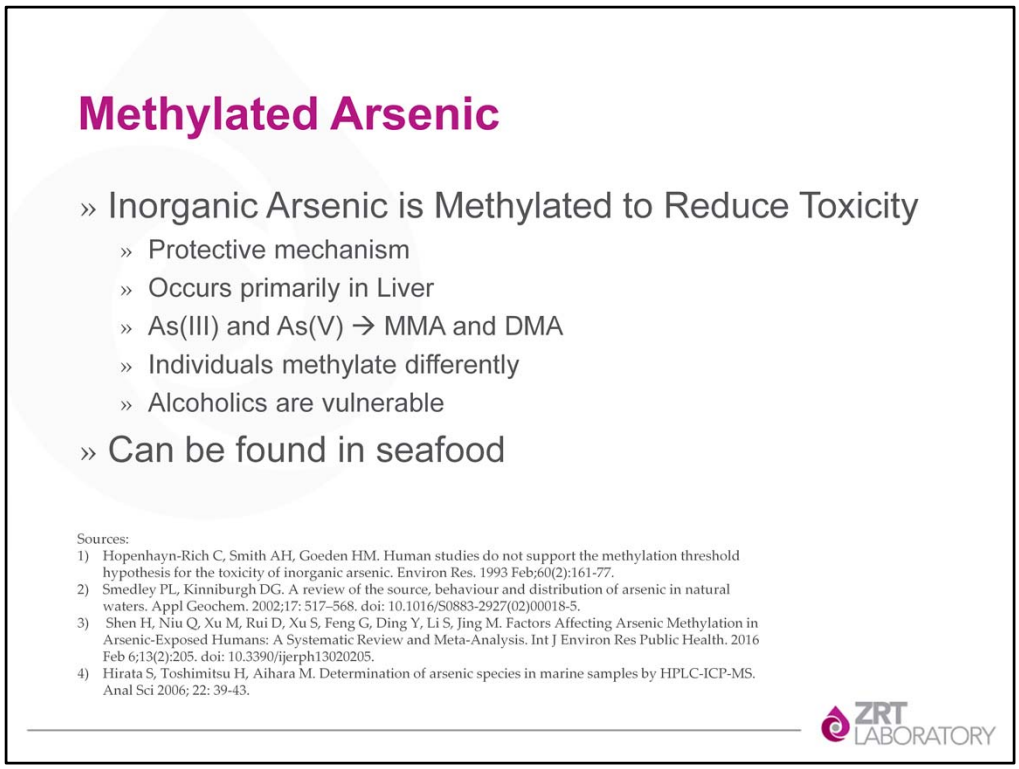
Chronic exposure to high levels of inorganic arsenic can lead to skin lesions, diabetes, hypertension, cardiovascular disease, neurological disorders, and many other ailments.

High intake has been linked to lung, prostate, bladder, renal, skin and numerous other cancers.

Inorganic arsenic will cross the placenta, but luckily protective mechanisms exist so that it is not transferred through breast milk.

The most common source of inorganic arsenic is private well water. It is very important that those with private wells have their water tested, as inorganic arsenic is odorless and colorless. Inorganic arsenic in water will contaminate food and drinks such as rice and apple juice.

Inorganic arsenic in the US diet ranges from around 1-20ug/day, but can vary widely depending on location.



Inorganic arsenic is methylated primarily in the liver to Monomethyl Arsenic and Dimethyl Arsenic. Methylated arsenic is not nearly as toxic as inorganic arsenic, but some methylated intermediates are potentially very dangerous.

After consumption of inorganic arsenic, around 10-15% will be methylated to monomethyl arsenic, 60-75% to dimethyl arsenic, and 10-20% remain unchanged.

Each person will metabolize arsenic differently, and it has been shown that certain populations have evolved to tolerate high levels of inorganic arsenic, specifically those living in Bangladesh and the Andes mountains where inorganic arsenic levels in water can reach 1000 parts per billion.

Alcohol consumption is know to affect arsenic methylation efficiency due to liver damage.

Methylated arsenic, primarily Dimethylarsenic, is commonly found in seafood along with high levels of organic arsenic.

Organic Arsenic

» Relatively Non-Toxic
» Primary source is seafood
» Travels through the body unchanged

Sources:
1) Andreae MO. Organoarsenic compounds in the environment. In: Craig PJ, ad. Organometallic compounds in the environment. Principles and reactions. New York: John Wiley, 1986:198-228.

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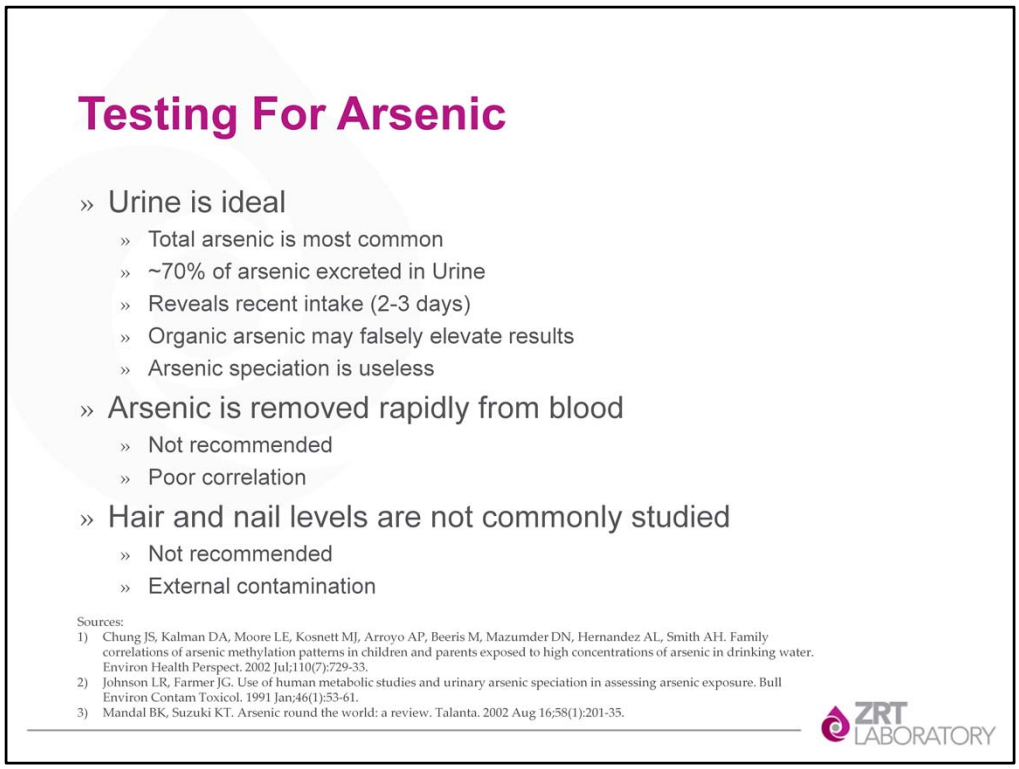
Organic arsenic is relatively non-toxic, and passes though the body unchanged in most cases.

The primary source of exposure is seafood. When fish or shellfish take up inorganic arsenic they convert it into organic and methylated arsenic species.

The two most common organic arsenic species are Arsenobetaine and Arsenocholine.



It is difficult to interpret urine arsenic results, whether they are speciated or not.

Total urine arsenic, which represents recent arsenic intake, will include inorganic, methylated, and organic arsenic species. The main species of interest in determining arsenic exposure is inorganic arsenic, as it is the most toxic. The problem is that inorganic arsenic is methylated in the body, then excreted, with very little inorganic arsenic left in it's original state. Seafood consumption will increase levels of organic and methylated arsenic which may falsely indicate elevated levels of inorganic arsenic consumption in both speciated and non-speciated testing.

Multiple studies have concluded that it is best to test for total arsenic in urine, and to try to identify the source of arsenic if urine levels are elevated, or to re-test if results are high, paying attention to the what was recently consumed. If seafood was consumed recently, that is most likely the culprit. If no seafood was consumed, then it would be best to test commonly used drinking water sources to identify if inorganic arsenic is present at elevated levels.

Arsenic is rapidly cleared from the blood, and studies have shown that blood arsenic does not correlate will with arsenic exposure from drinking water while urine arsenic correlates well. The only time blood arsenic should be used is as an indicator of recent acute exposure.

Hair and nail arsenic levels are rarely used to determine arsenic exposure, and they are

prone to external contamination.

Lead

» #2 on the ATSDR Priority List
» IARC Probable Human Carcinogen
» Long Half Life of >25 Years in Bone
» Just Realizing Low Level Toxic Effects in Children

Sources:
1) IARC. 2006. Inorganic and organic lead compounds. In: IARC monographs on the evaluation of carcinogenic risks to humans. Lyon, France: International Agency for Research on Cancer. p. 519.
2) Agency for Toxic Substances and Disease Registry (ATSDR).Toxicological profile for lead. Atlanta (GA): US Departmentof Health and Human Services, Public Health Service; 2007.

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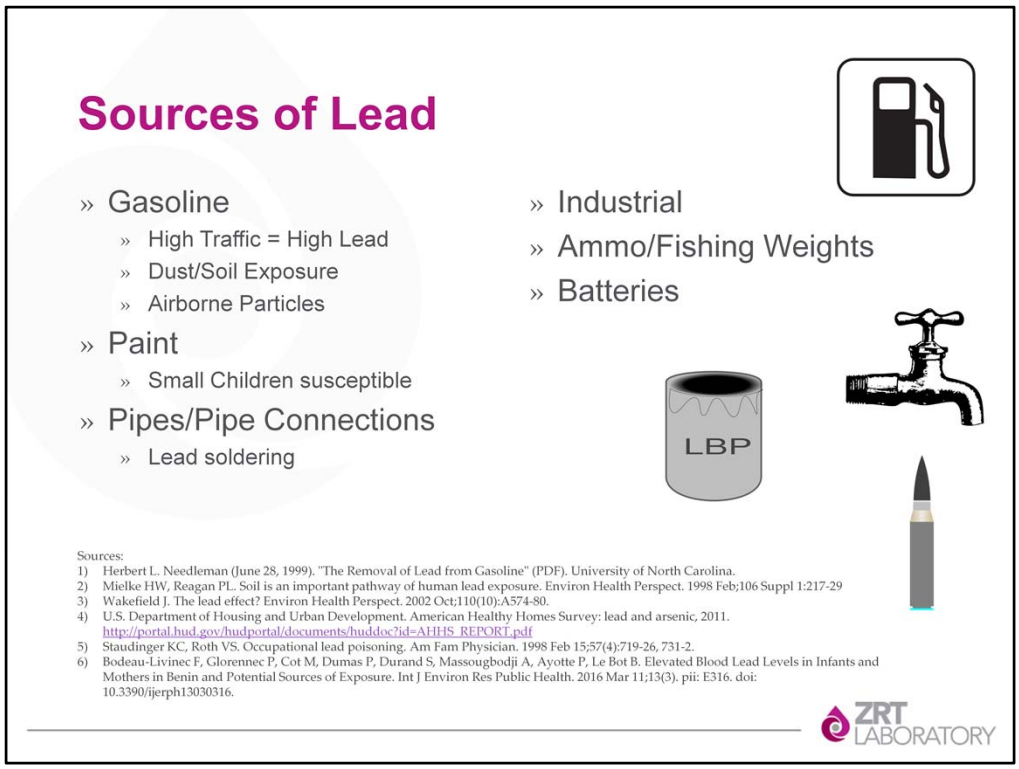
Now we move on to lead.

Lead has been all over the news recently. Flint, Michigan and currently my home town of Portland, Oregon have identified lead problems that, in reality, exist throughout the United States.

Lead is #2 on the A T S D R's hazardous substances priority list, primarily because of its wide spread use over the last century and its detrimental neurotoxic effects, particularly in children.

Lead mimics calcium ions, which allows it to get into bones and other organs, resulting in a long half life of over 25 years. When bone breaks down, it releases lead into the blood stream, resulting in a reoccurring source of exposure.

New research is beginning to show the toxic effects of lead, particularly in children, at levels much lower than we once thought were toxic.



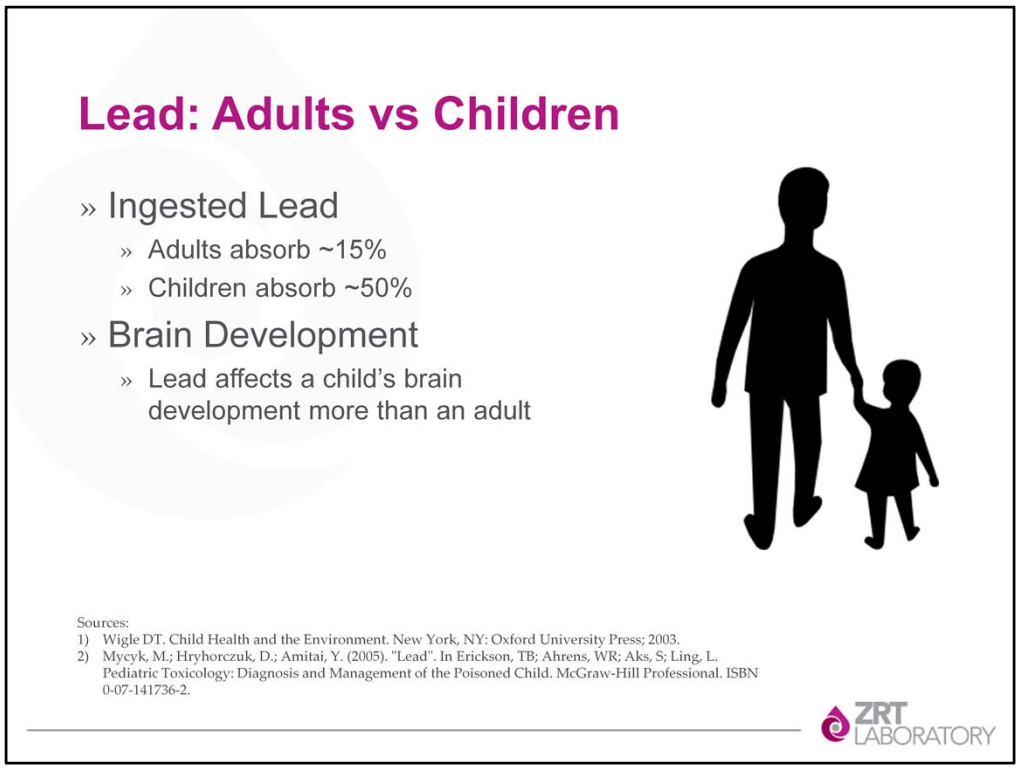
There are many sources of lead exposure.

When leaded gasoline was first introduced in the 1950's, atmospheric lead pollution increased dramatically. Around the same time, the use of leaded paint peaked and was present in nearly every home. It is interesting to note that levels of lead in city dust correlate well to historical traffic flow volumes when lead was present in gasoline.

The CDC estimates that currently around 24 million houses have deteriorating lead paint in the United States, which is around 1 in 4 houses. Even a small paint chip can contain hundreds of milligrams of lead.

The National Institute of Occupational Safety and Health estimates that around 3 million workers are exposed to lead in the workplace, and many employees bring it home on their shoes and clothes.

Lead used in ammunition can cause harm to an animal or human if a lead pellet or fragment of a lead pellet is consumed. Also, if a bullet is lodged in the body and not removed, it can be a significant source of lead exposure.



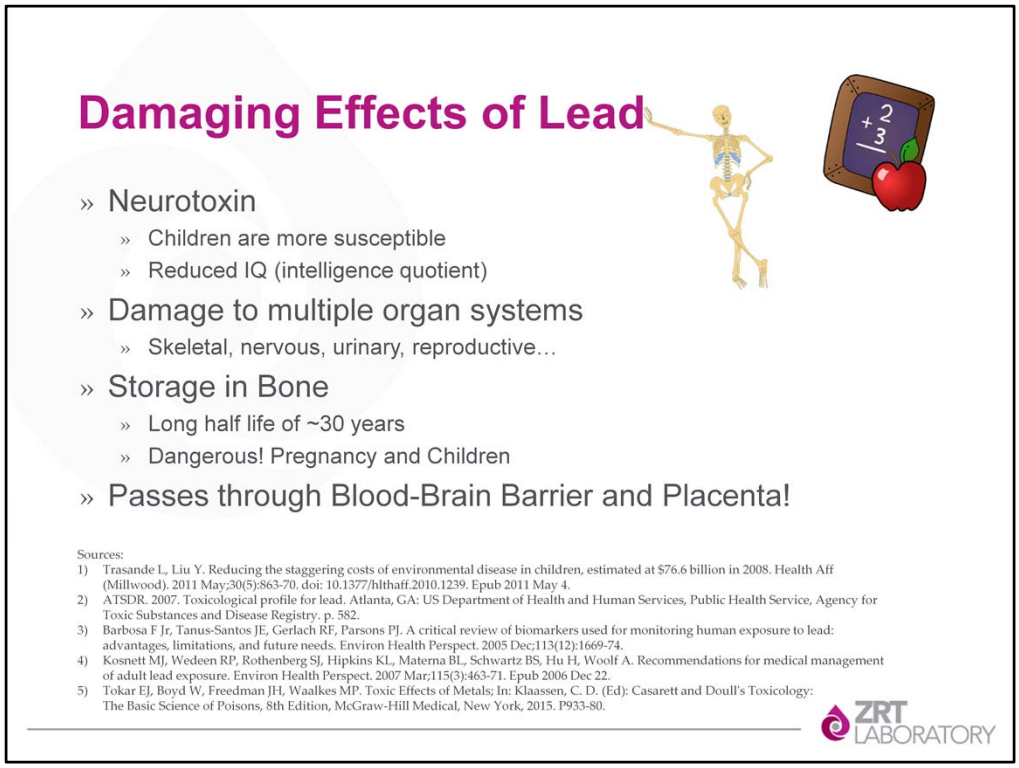
Lead affects children and adults differently.

Ingested lead is well absorbed in children at around 50%, while in adults it is only around 15%. This can be made worse if nutrient deficiencies exist. This will be touched on a bit later.

Lead is most detrimental during the first years of life. It interferes with the organization of ion channels, synapse formation, and neurochemical development in a child's developing brain. This is less pronounced in adults.

Lead is dangerous for two primary reasons, the generation of reactive oxygen species and the depletion of antioxidant reserves.

Lead, like other heavy metals has a strong affinity for sulfur and selenium. The sulfhydryl group on glutathione, a very important antioxidant, will bind directly to lead, effectively inactivating the glutathione molecule. The same goes for the selenium in glutathione peroxidase.



Why is lead so dangerous?

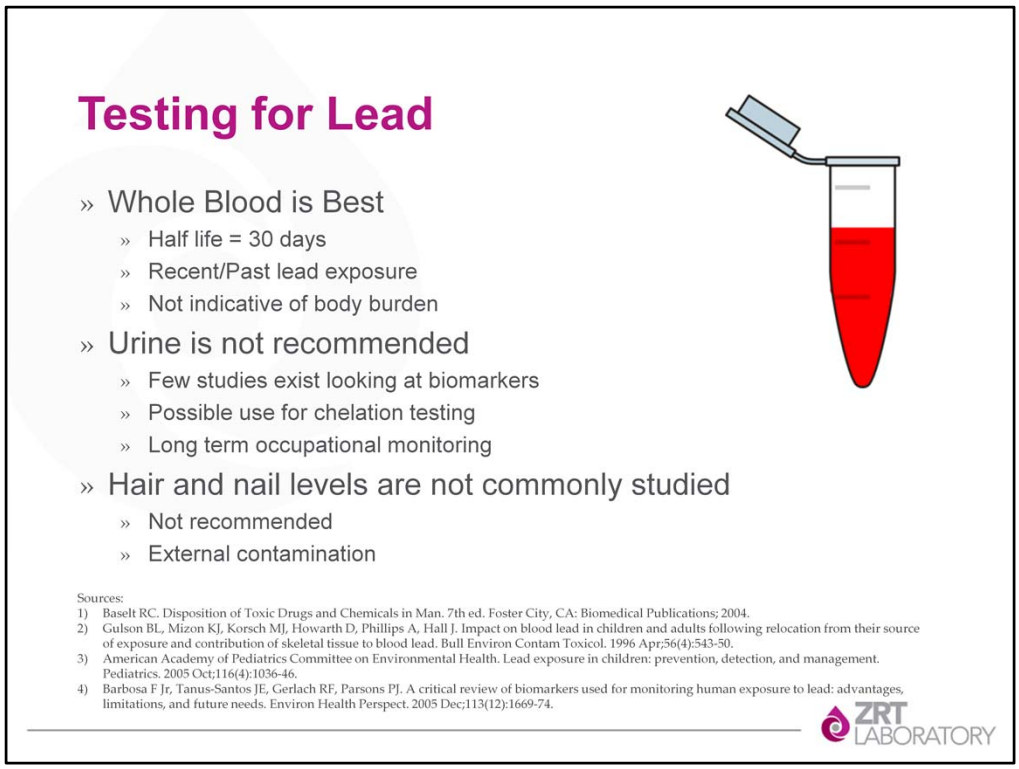
It has been estimated that the economic loss due to childhood lead poisoning is around $61 billion US dollars per year.

Lead is a potent neurotoxin that sticks around in the body for a very long time. Lead has been associated with adverse health effects on multiple organ systems, including the skeletal, nervous, urinary, cardiovascular, immune, gastrointestinal, and reproductive systems. The brain is the organ most sensitive to lead exposure.

Some of the effects of lead poisoning include headaches, agitation, delayed reaction times, irritability, muscle weakness, numbness and tingling, sleep issues, slurred speech, anemia, along with many other ailments.

Around 96% of the lead that is absorbed by the body will be stored in bone. The half life of lead in bone is around 30 years, meaning that it will be released and reabsorbed when bone breaks down. This is especially a concern for pregnant women and children, as lead is released during formation of a fetal skeleton and during bone growth.

Lead also has the ability to pass through the blood-brain barrier and across the placenta by substituting for calcium ions in Calcium-ATPase pumps.



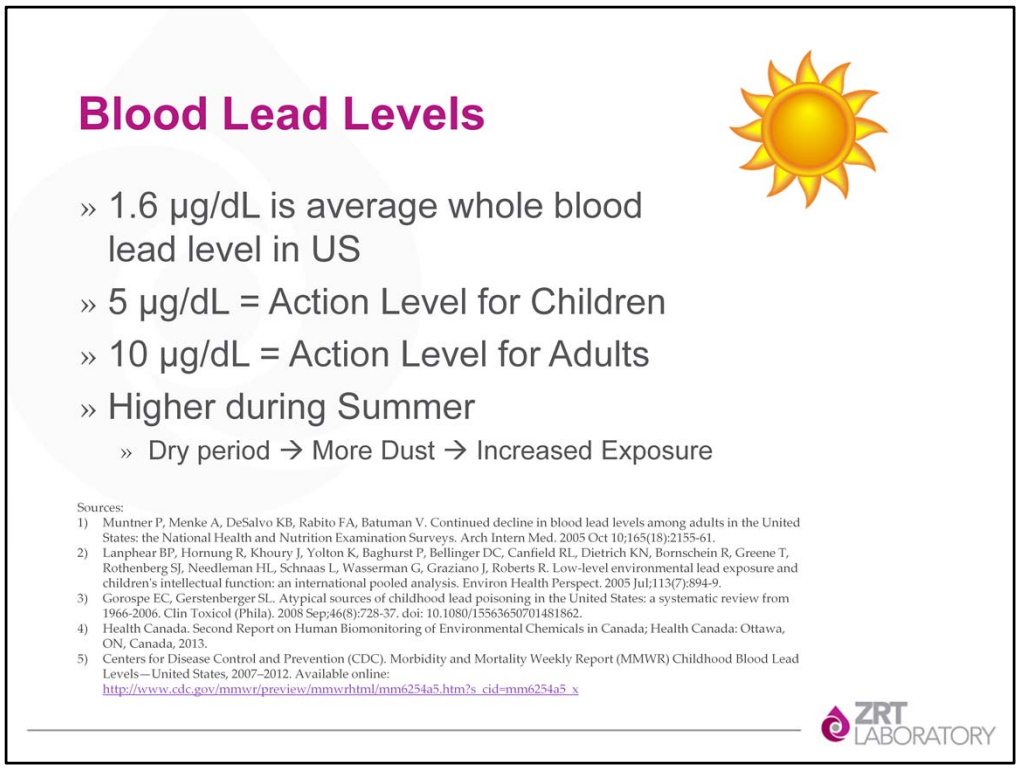
Testing for Lead.

Lead is most commonly tested via whole blood, whether it be from a capillary finger prick or venous puncture, because 95% of circulating lead is bound to red blood cells. Serum is not an ideal sample choice.

The half life of lead in blood is around 30 days, which is about the same as the life-span of a red blood cell. Lead that is mobilized from the bone back into blood can cause a significant increase in blood lead levels, especially in children where up to 90% of blood lead can be from release during bone growth.

Few studies exist that look at the comparison of urine lead levels to biomarkers of exposure. Urine lead levels may be useful for monitoring chelation or for long term occupational monitoring.

Just like arsenic, monitoring lead levels in hair and nails is not recommended due to possible external contamination and lack of research.



What blood lead levels should you expect to see after testing, and when should action be taken?

First off, there is no safe level of lead exposure. Action levels for blood lead were once at 100 ug/dL, yet have been dropping lower and lower due to new research. Recent studies have shown that behavioral and neurological effects can occur at blood lead levels below 5 ug/dL in children. It has been estimated that if a child's blood lead level raises from less than 1 ug/dL to 10 ug/dL, they will lose around 6 IQ points.

In the United States, a recent NHANES study showed that average blood lead level is 1.6ug/dL, yet around 450 thousand children have blood lead levels >5 ug/dL.

The removal of lead from gasoline and paint resulted in a reduction of US childhood blood lead levels from 15 ug/dL in the 1970's to 1.8ug/dL in 1999.

It is interesting to note that during the summer months, blood lead levels will rise due to increased lead exposure from contaminated dust; a result of dry conditions.

Mercury

» #3 on the ATSDR Priority List

» Well known and researched heavy metal

» 50-75% of environmental mercury comes from anthropogenic sources

Sources:
1) USGS (United States Geological Survey). 2009. Mercury in fish, bed sediment, and water from streams across the United States, 1998-2005. Scientific Investigations Report 2009-5109.
2) Davidson PW, Myers GJ, Weiss B. Mercury exposure and child development outcomes. Pediatrics. 2004 Apr;113(4 Suppl):1023-9.

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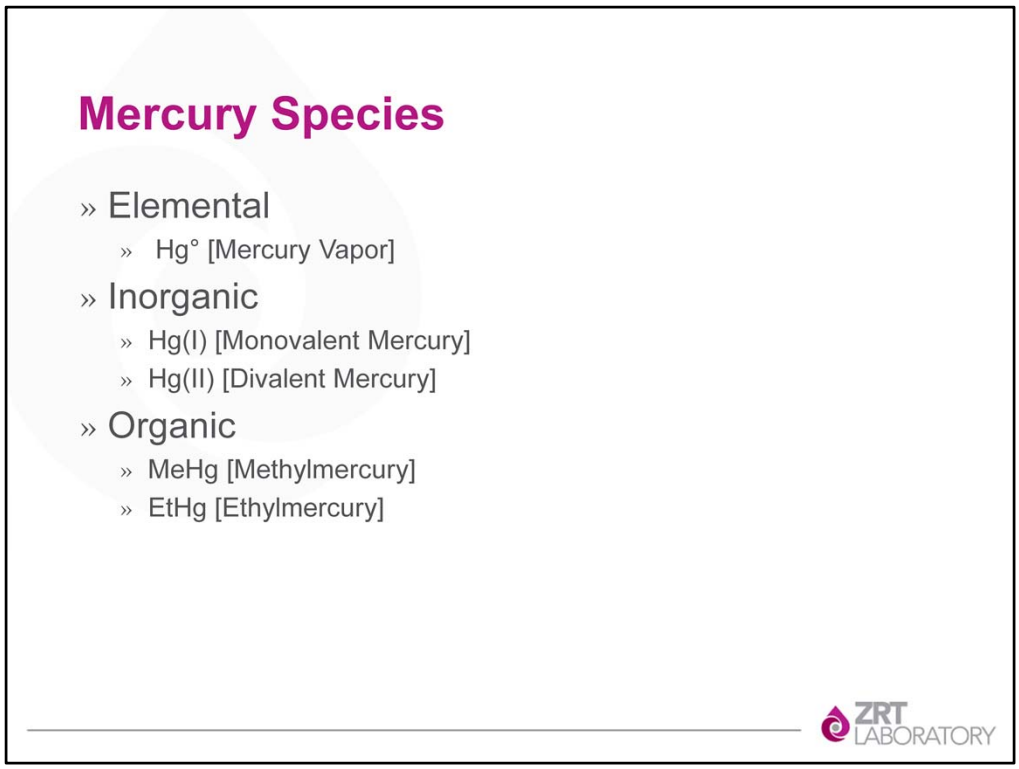
Third on A T S D R ’s priority list is Mercury.

Mercury is a well known heavy metal that is commonly linked to vaccines, dental amalgams, coal combustion, and fish consumption.

Sadly, a majority of the mercury in our environment comes from human sources.



First we will start by going over the three unique mercury species. The level of mercury toxicity is dependent on not only the species, but how it enters into, or is processed by the body.

Elemental mercury, or what is commonly called quicksilver, is what most people think of when they hear the word mercury. It is the only metallic element that is a liquid at standard conditions for temperature and pressure.

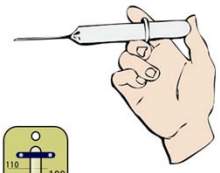
Inorganic mercury can come from a wide variety of sources, but is also formed from elemental and organic mercury once in the body.

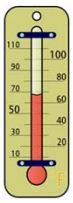
Organic mercury is the most toxic form, which comes primarily from fish and vaccinations.

We will dig a bit deeper into each species of mercury in the next slides.

Elemental Mercury

- Source
  - Dental amalgams, thermometers, gold-mining, laboratory
- Absorption
  - ~80% via inhalation (readily vaporizes)
  - ~0.01% via ingestion
  - Lipid Soluble (Crosses Blood-Brain/Placental Barrier)
- Excretion
  - Urine, feces, sweat, saliva
- Transportation
  - Binds to sulfhydryl groups
  - Suspended in plasma
- Transformation
  - Rapidly oxidized to inorganic mercury





Sources:
1) WHO (World Health Organization). 2003. Elemental mercury and inorganic mercury compounds: Human health aspects. Concise International Chemical Assessment Document 50.
2) Ballatori N, Clarkson TW. Biliary secretion of glutathione and of glutathione-metal complexes. Fundam Appl Toxicol. 1985 Oct;5(5):816-31.
3) Friberg L, Mottet NK. Accumulation of methylmercury and inorganic mercury in the brain. Biol Trace Elem Res. 1989 Jul-Sep;21:201-6.

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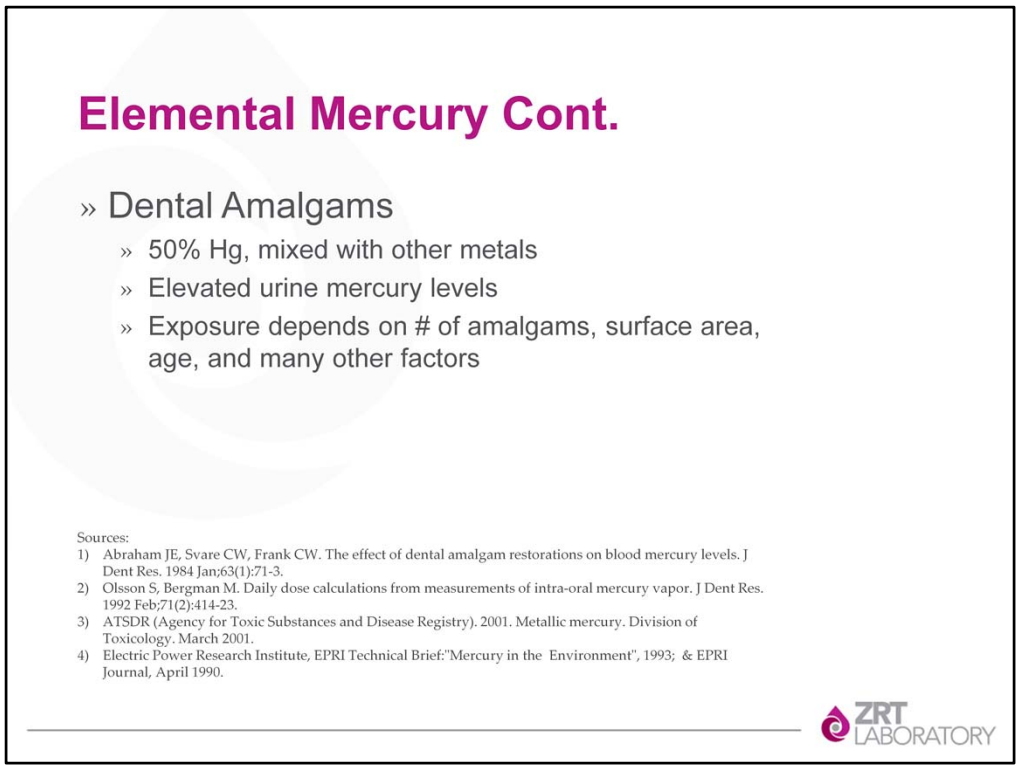
Elemental mercury is the species that most people are familiar with. It was present in science classes of the past, is used in thermometers, and is the main component of dental amalgams.

While elemental mercury is a liquid at room temperature, it is constantly vaporizing. Heating mercury will speed up the rate at which mercury vaporizes. Vacuum or ventilation systems can spread mercury vapor, which is heavier than air and remains close to the floor, a particular danger to crawling babies.

As elemental mercury is inhaled, around 80% of it is absorbed. It enters the blood stream and binds to sulfur on red blood cells, glutathione, or metallothionein, or is transported suspended in plasma. Even though elemental mercury is rapidly oxidized to inorganic mercury, some free elemental mercury will cross the blood brain barrier before being oxidized, resulting in mercury that is essentially unable to cross back out of the brain due to its ionic charge.

Elemental mercury is excreted primary through urine and feces after conversion to inorganic mercury.

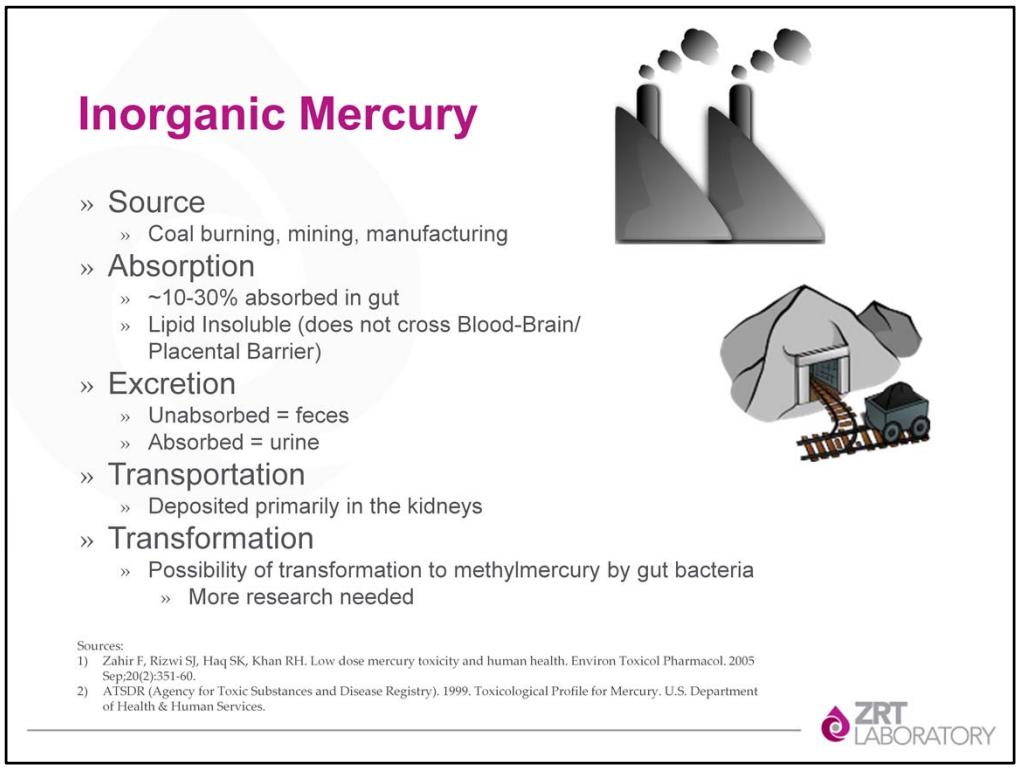


Dental amalgams are a major source of elemental mercury exposure.

Dental amalgams are made up of around 50% mercury and a mix of other metals such as copper, tin, silver and zinc. Often called silver fillings because of their color, many people are unaware of the amount of mercury being placed in their mouth. The most common alternate to dental amalgam fillings are composite fillings, but they have a shorter life span and are more expensive. Mercury waste from dental offices is a huge problem, as it gets into waste water which is then transferred throughout the environment.

The average mouth of someone with dental amalgams contains 2.5 grams of mercury. 0.5 grams of mercury is enough to contaminate a 10 acre lake to the point where the government would recommend that fish are not consumed.

The amount of exposure to mercury vapor or particles from dental amalgams depends on the number of amalgams, age, tooth brushing, diet, oral breathing habits, body weight and numerous other factors. Recent studies have shown that electromagnetic waves from objects like cell phones can increase the amount of mercury gassing off of amalgams.

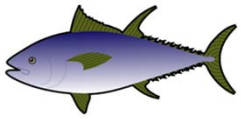


Inorganic mercury can come from a wide variety of anthropogenic sources. Coal fired power plants are the largest source of airborne mercury. When inorganic mercury is deposited in our ecosystem, bacteria can convert it into extremely toxic organic mercury.

Inorganic mercury is not absorbed well when ingested, which is the most common route of exposure. Once absorbed it eventually makes its way to the kidneys were it accumulates bound primarily to sulfhydryl groups.

A few researchers believe that inorganic mercury can be converted to organic mercury by gut or oral bacteria, but more research is needed.

Organic Mercury



- Source
  - Fish! (can be higher in freshwater than saltwater)
    - Not all fish are the same
  - Vaccinations
- Absorption
  - ~95% in GI tract
  - Lipid Soluble (Crosses Blood-Brain/Placental Barrier)
- Excretion
  - >90% in feces
  - ~10% in urine as inorganic mercury
- Transportation
  - Readily crosses blood-brain barrier and placenta + in breast milk
  - Binds to a L-Cysteine complex, transported via amino acid carriers into brain
- Transformation
  - Slowly de-methylated to inorganic mercury = Big Problem!!!

Sources:
1) USEPA (United States Environmental Protection Agency). 2008. Initial risk-based prioritization of mercury in certain products. Risk-Based Prioritization Document. November 2008.
2) Ullrich SM, Tanton TW, Abdrashitova SA. Mercury in the aquatic environment: a review of factors affecting methylation. Crit Rev Environ Sci Technol. 2001;31(3):241–293.
3) ATSDR (Agency for Toxic Substances and Disease Registry). 1999. Toxicological Profile for Mercury. U.S. Department of Health & Human Services.
4) WHO (World Health Organization). 2010. Children's exposure to mercury compounds. ISBN 978 92 4 150045 6.
5) Food and Drug Administration. Food safety [Web page]. www.fda.gov/Food/FoodSafety.

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Organic mercury is the most toxic of the three species.

Inorganic and elemental mercury spread throughout our environment can be converted to methylmercury by bacteria. Due to low oxygen content at the bottoms of rivers and lakes, and the presence of sulfate reducing bacteria, there is a high amount of mercury methylation from inorganic to organic.

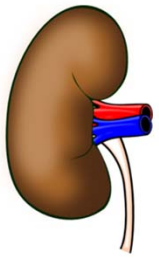
Organic mercury makes its way through the food chain with higher predatory fish containing the most mercury. In most cases over 90% of the mercury present in fish tissue is methylmercury. Large fish such as tuna and shark may contain as much as 500 parts per billion mercury in edible tissue.

Organic mercury is highly absorbed in the gastrointestinal tract, with around 10% ending up in the brain and the remainder distributed throughout the body. It will also cross the placental barrier, exposing a fetus to mercury.

Organic mercury will eventually de-methylate and become locked into organs such as the brain because of its ionic charge. This results in the body accumulating a large amount of mercury, which will bind primarily to sulfur and selenium, inactivating enzymes, antioxidants, and individual elements.

Inorganic mercury has a half life in the body of 20 years.

Mercury Toxicity



- High affinity for Sulfur/Selenium
  - Binds to cysteine = inactivates cysteine enzymes/proteins
  - Binds to selenium = cannot be used
    - Reduces toxicity, which is good
- Symptoms
  - Tremors, irritability, peripheral neuropathy, vision changes, hearing loss, kidney problems

Sources:
1) ACGIH (American Conference of Governmental Industrial Hygienists). 2001. Documentation of Biological Exposure Indices. 7th edition. Mercury, elemental and inorganic. ACGIH Press, Cincinatti, OH.
2) WHO (World Health Organization). 2010. Children's exposure to mercury compounds. ISBN 978 92 4 150045 6.

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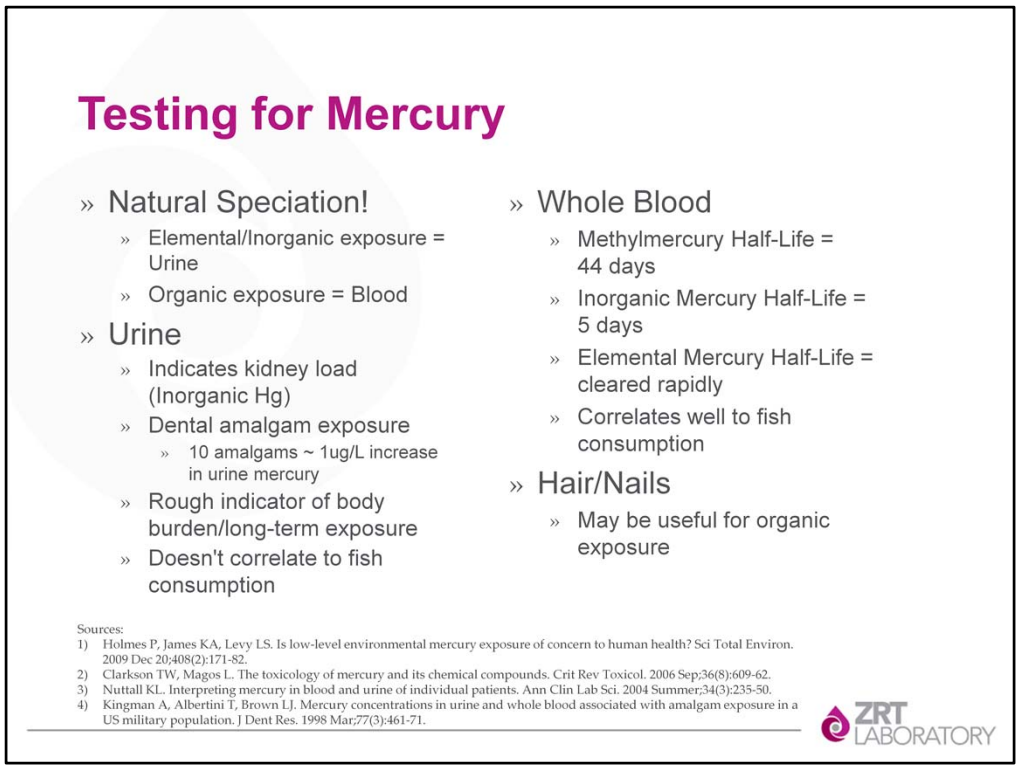
So, what does mercury toxicity look like?

Mercury toxicity can present itself in many different ways. One of the main reasons mercury is toxic is because of it's high affinity for selenium and sulfur, both of which are present in essential antioxidants and enzymes. In many cases symptoms will only be present after very high exposure and will vary from person to person. Low level mercury exposure is still dangerous even without symptomatology.

Some common symptoms associated with mercury toxicity are numbness and tingling, kidney issues, hearing problems, and irritability.



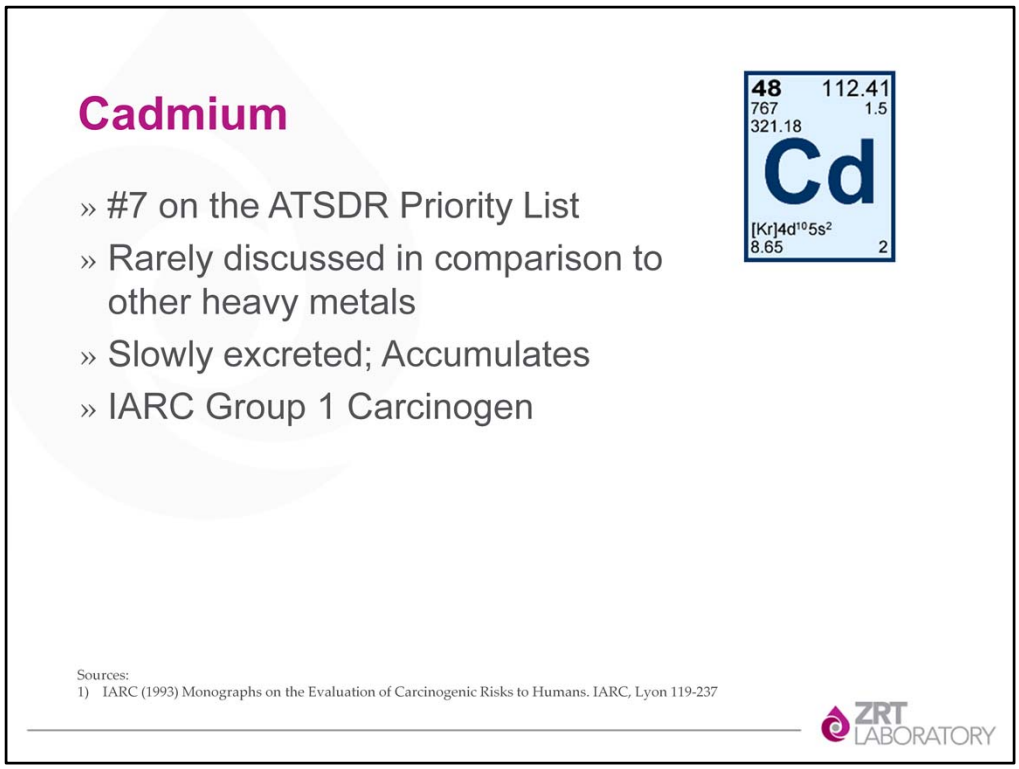
How do you test for mercury, and what exactly do the results mean? Testing both urine and blood provides a full picture of mercury exposure and takes advantage of natural speciation by the body.

The most reliable indicator of long term inorganic and elemental mercury exposure is urine, as these two species are quickly cleared from blood and accumulate in kidney tissue. Whole Blood on the other hand is the best indicator of organic mercury exposure due to its long half-life bound to red blood cells.

Studies have show good correlations between urine mercury levels and the number of dental amalgams. For every 10 amalgams, urine mercury will raise around 1 ug/L. Strong connections have also been made between fish consumption and blood mercury levels. High blood mercury levels are likely when fish is a large part of a diet, which is commonly seen in Asian and Scandinavian countries.

It is important to note that neither blood or urine is a useful indicator of total body burden. In order to assess this, one would require tissue analysis of the mercury tightly bound throughout the body.

Hair and nail mercury may be a good indicator of long term organic mercury exposure, but is prone to external contamination. Hair and nail mercury levels do not correlate well with inorganic or elemental exposure but sometimes do with fish consumption if it is a long term dietary staple.



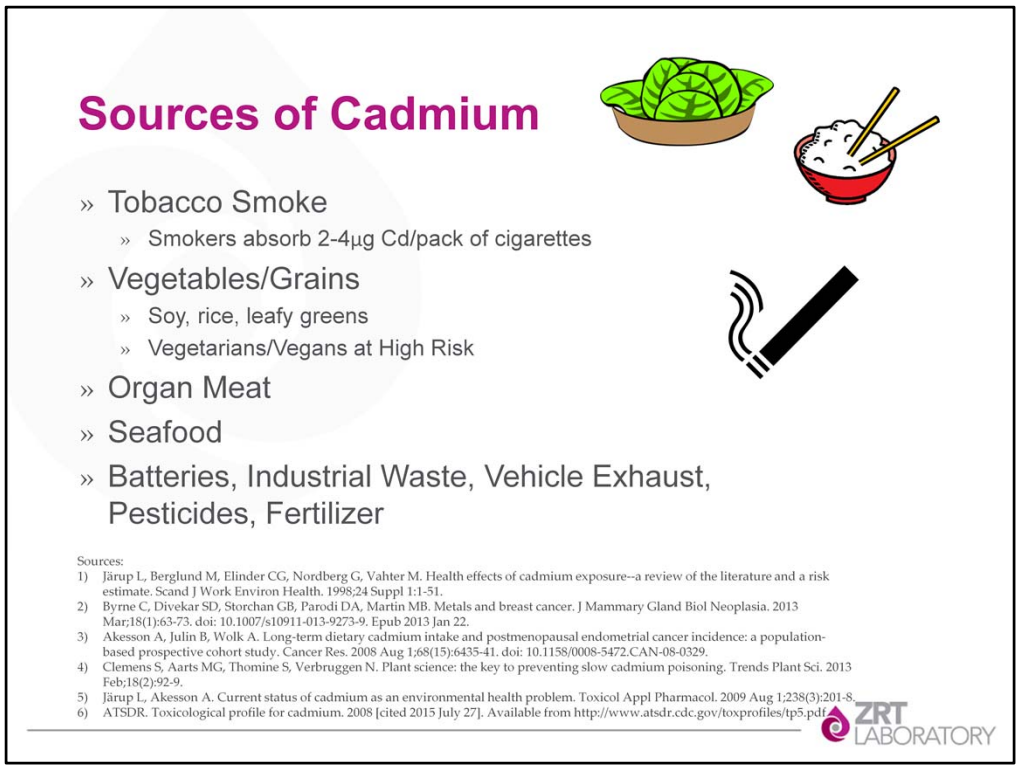
Cadmium is the last of the Big 4 heavy metals I will discuss today. It is #7 on the A T S D R hazardous substances priority list.

It is rarely discussed in comparison to the other major heavy metals, but may be one of the most dangerous.

Once inside the body, cadmium has a very long half-life and will bioaccumulate because it is excreted incredibly slow.

Although some health agencies have cadmium listed as a potential carcinogen, the International Agency for Research on Cancer has listed it as a group 1 carcinogen.

Recently in Portland, Oregon, we have had issues with cadmium pollution from a glass factory, resulting in chronic exposure and possible breast cancer clusters surrounding the factory.



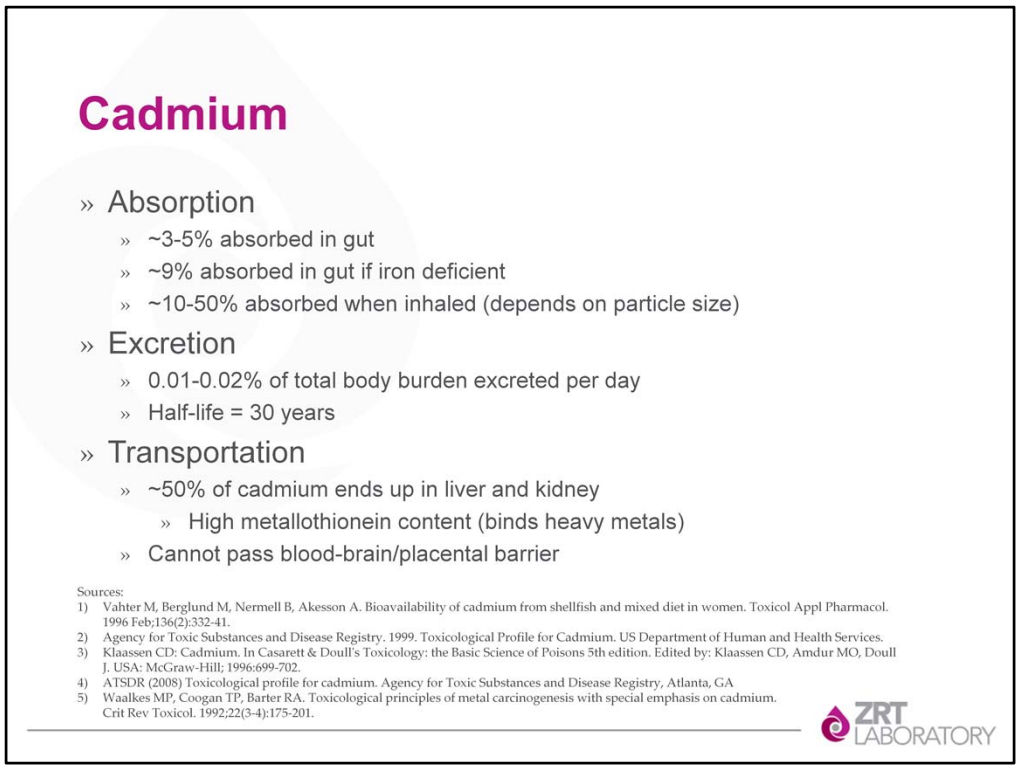
There are many sources of cadmium exposure, but the primary source for those who don't smoke tobacco is the food we eat.

Smokers absorb around 1-3µg of cadmium per day, which is around the same amount absorbed from a non-smokers diet. Studies have shown that smokers have blood cadmium levels around 3 times higher than non-smokers.

Cadmium is naturally found in soil, but can also be deposited on soil from industrial waste, vehicle exhaust, and fertilizers to name a few examples. Plants take up cadmium from soil, with some accumulating it at higher levels than others. Tobacco, for example, is very good at accumulating cadmium. Rice, soy, and leafy greens are also good at accumulating cadmium, and are dietary staples for many populations.

Organ meat can be very high in cadmium, specifically liver and kidney where cadmium is known to accumulate due to metallothionein production.

Interestingly, women found to have the highest levels of urinary cadmium were non-smokers, have a high level of education, and consume twice as many vegetables and whole grains than those with the lowest levels of urinary cadmium. Multiple studies have shown that vegetarians and vegans have a much higher intake of cadmium due to increased grain and vegetable consumption along with dietary nutrient deficiencies. One study showed that vegetarians had blood cadmium levels 3 times higher than non-vegetarians.

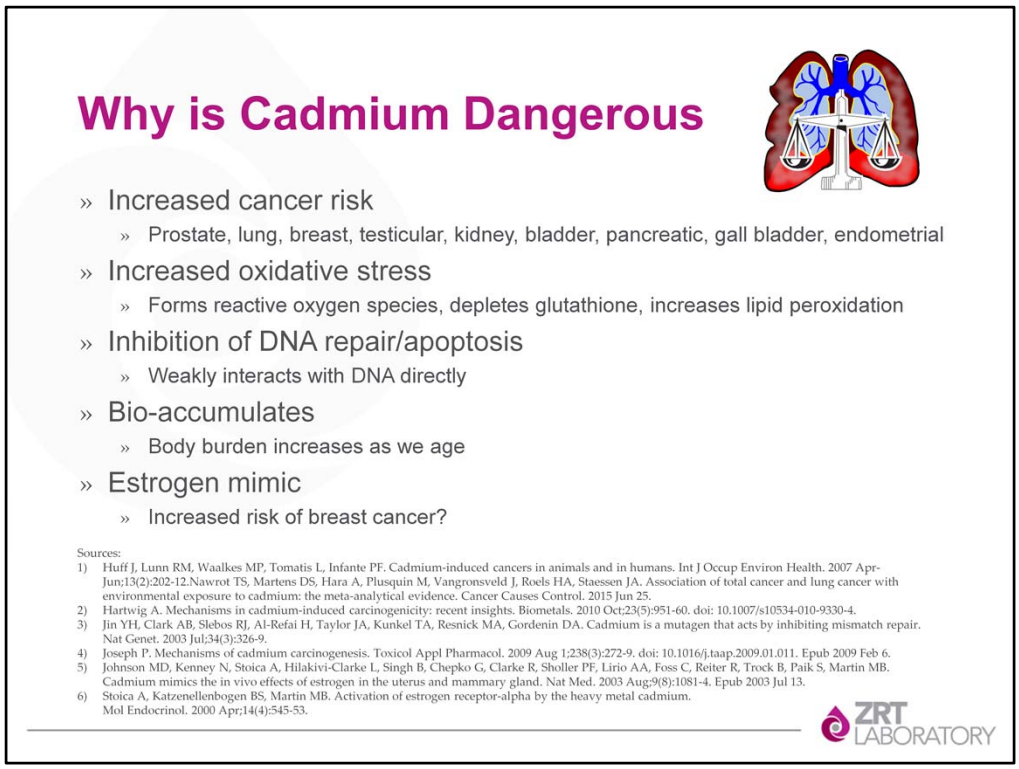


Cadmium is poorly absorbed in the gut. Humans consume around 8 to 30ug of cadmium a day, but only around 3-5% is absorbed. Inhalation of cadmium is much higher at 10 to 50%, which depends primarily on particle size. Because of this, smokers will take in around double the cadmium of non-smokes every day.

Cadmium is excreted from the body very slow. Only about 0.01 to 0.02% of the total body burden is excreted each day, with a half-life of around 30 years. This results in a significant increase in cadmium body burden as we age.

As cadmium is absorbed from inhalation or ingestion, it enters the blood stream and can be found in red blood cells. Around 50% of absorbed cadmium ends up in the liver and kidneys due to the high metallothionein content, a detoxifying protein that will be discussed later.

Cadmium luckily has a hard time crossing the blood-brain and placental barrier.



So, why is cadmium dangerous?

Cadmium exposure has been linked to numerous types of cancer, including prostate, lung, breast, testicular, kidney, bladder, pancreatic, gall bladder, and endometrial, and is a group 1 carcinogen established by the International Agency for Research on Cancer.

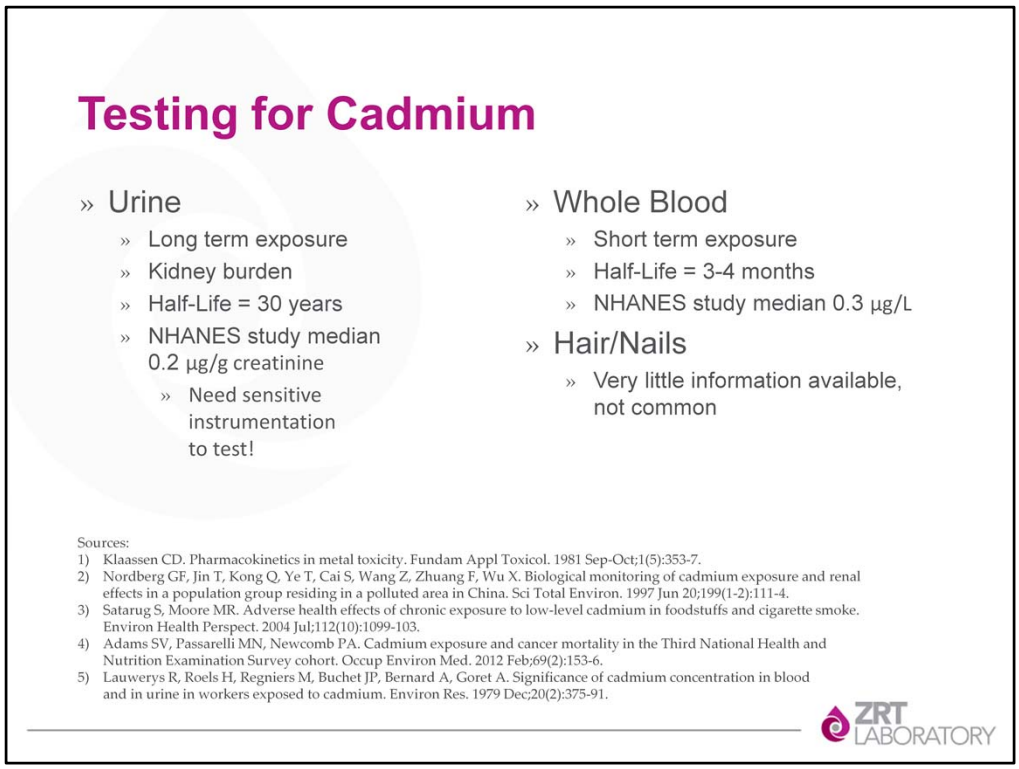
A study of over 20000 US and Belgian participants found that each doubling of urinary cadmium resulted in a pooled estimate relative risk increase of 22% for all cancers and 68% for lung cancer.
Most studies have shown that very small increases in cadmium body burden will significantly increase the possibility of cancer developing. Just like lead, very low levels of exposure are proving to be much more damaging than previously expected.

Like other heavy metals, it is believed that cadmium is most damaging because of increased oxidative stress. Cadmium will form reactive oxygen species, deplete glutathione and other sulful/selenium containing antioxidants, and increases lipid peroxidation.

Cadmium weakly interacts with DNA, so it is believed that its toxic effects against DNA may be through epigenetic or other mechanisms such as inhibition of DNA repair and apoptosis.

Cadmium will accumulate due to its slow excretion. Newborns have negligible cadmium burden, but by age thirty the body will have accumulated around 30 to 50mg of cadmium.

Cadmium shares similar properties with estrogens, allowing it to bind to estrogen receptors and form high-affinity complexes. This could be one of the mechanisms behind the increased risk of breast cancer with higher cadmium intake. A US study showed that women with urine cadmium levels above 0.58 ug/g creatinine had twice the breast cancer risk as those below 0.26ug/g creatinine. If you are interested more in this topic, my father Dr. David Zava and I did a webinar specifically on cadmium and breast cancer this past year, which can be found on our website.



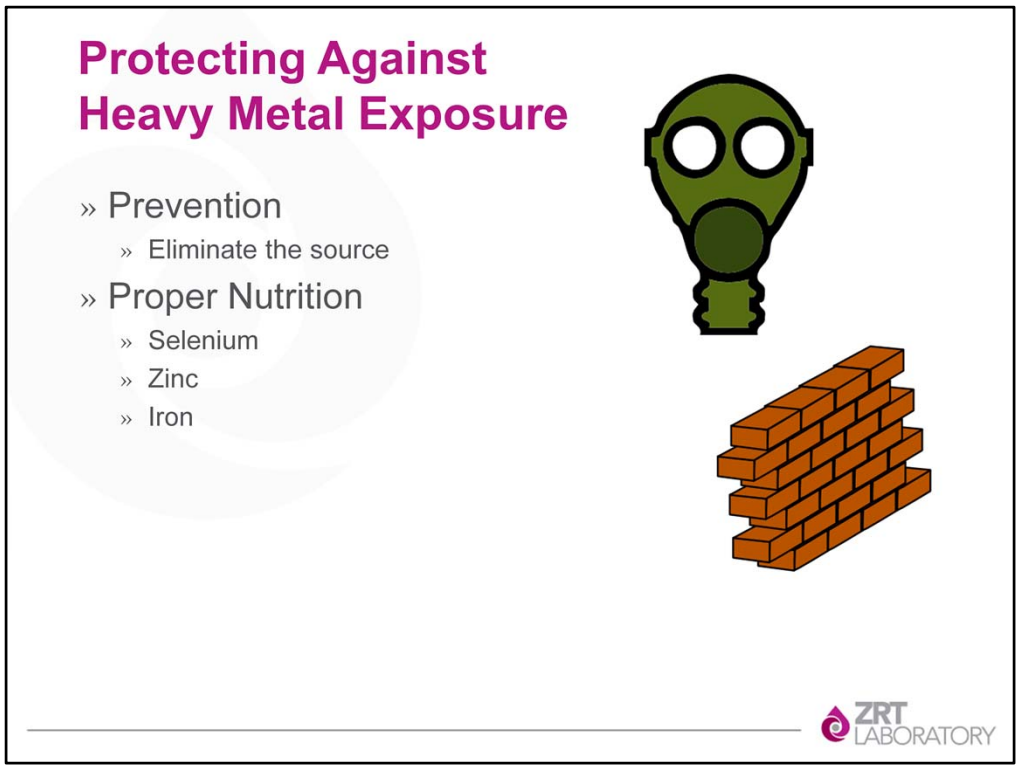
Testing for cadmium can be very difficult. Very advanced element analysis machinery is required due to greater than 95% of results testing in the parts per trillion.

The most recent National Report on Human Exposure to Environmental Chemicals, or NHANES, showed a median urine cadmium level of 0.2 ug/g creatinine and median blood cadmium level of 0.3 ug/L for 20+ year olds. The European Union has proposed that urinary cadmium should fall below 0.66ug/g creatinine, reflecting recent findings on adverse effects of low-level cadmium exposure. The World Health Organization set a urine cadmium threshold at 5.24 ug/g creatinine, while the European Food Safety Authority set their action level to 1 ug/g creatinine. Urine cadmium levels around 2.5 ug/g creatinine have been shown to induce tubular damage in the kidneys. This shows the discrepancy between different groups as to what level of exposure is considered dangerous. Regardless, sensitive testing is needed to determine exact cadmium concentrations into the parts per trillion to assess cadmium exposure.

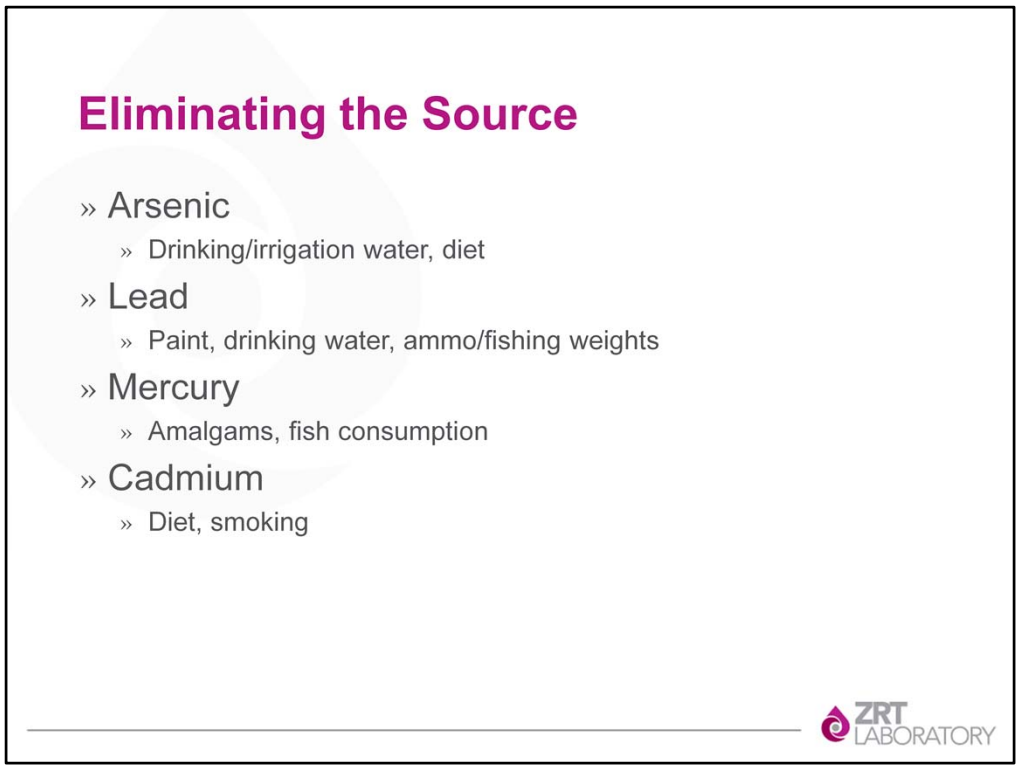
Urine is the best indicator of long term cadmium exposure and kidney burden. Urine will indicate exposure that has occurred over the last 30 years.

Blood cadmium is the best indicator of recent cadmium intake due to its short half-life of 3 to 4 months, around the same life span as a red blood cell. Urine cadmium will not increase significantly after acute exposure, but blood cadmium will show a near immediate increase. There is little use for urine testing during the first year of exposure.

For example, if someone smoked cigarettes habitually 5 years ago, their blood cadmium may show low while their urine cadmium will be high. On the other hand, if someone recently starts smoking cigarettes, their urine cadmium may be low, but their blood cadmium will be elevated.



At this point you are probably wondering how to protect against dangerous heavy metals. We will briefly discus how to prevent exposure and the benefits of proper nutrition in the fight against heavy metal exposure.



Here is a list of primary sources of exposure for the 4 major heavy metals. If a patient tests high for any of these metals in blood or urine, it is important to eliminate the source of exposure before moving forward with any treatment. In some cases, once exposure has occurred, the only thing that can be done is to prevent further exposure.

Understanding the half-life of each metal in different sample types will help assist in determining if exposure is current or from the past.

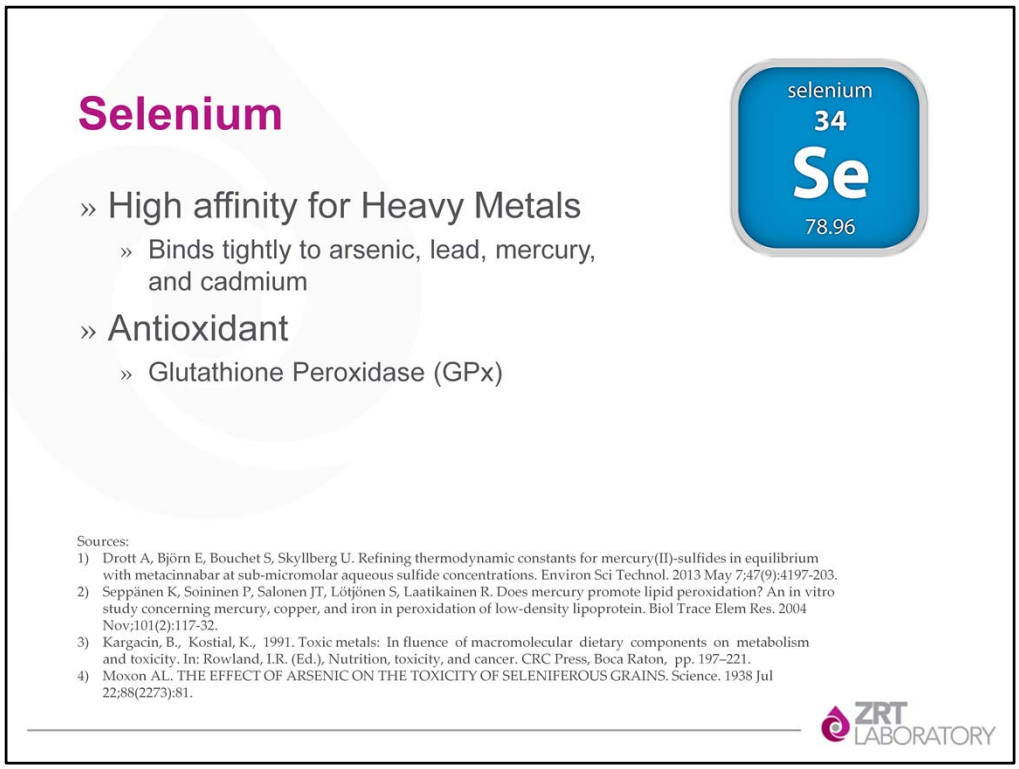
The most common source of arsenic exposure is from contaminated or naturally high drinking and irrigation water. Chronic arsenic exposure is common for those with untested well water. Arsenic removal filters or water treatment can help prevent this.

The major sources of lead exposure are paint, drinking water, and leaded ammo and fishing weights. Many cities provide free water and paint testing. How old is your home? If it was built prior to the 1970's, there is a high chance of leaded paint and pipes. Hunters and fisherman may be exposed to lead due to accidental consumption of shotgun pellets or bullets, or touching of hand to mouth after handing lead weights.

Two major sources of mercury are fish consumption and dental amalgams. How often do you eat fish, what are the serving sizes, and what type of fish are you consuming? Many online resources will help you make good decisions on what type of fish has the lowest mercury content. Dental amalgam removal is becoming a popular option to reduce mercury exposure. Currently we are validating a test for saliva mercury to help patients determine

how much mercury they are exposed to.

Cadmium exposure occurs primarily through diet and smoking. If you are a vegan or vegetarian, make sure that you are eating a well balanced diet to increase antioxidant levels and prevent nutrient deficiencies, as cadmium intake is much higher due to increased consumption of vegetables and grains. If you are a smoker, cessation of smoking will reduce cadmium intake by around half, reducing cadmium accumulation and body burden.



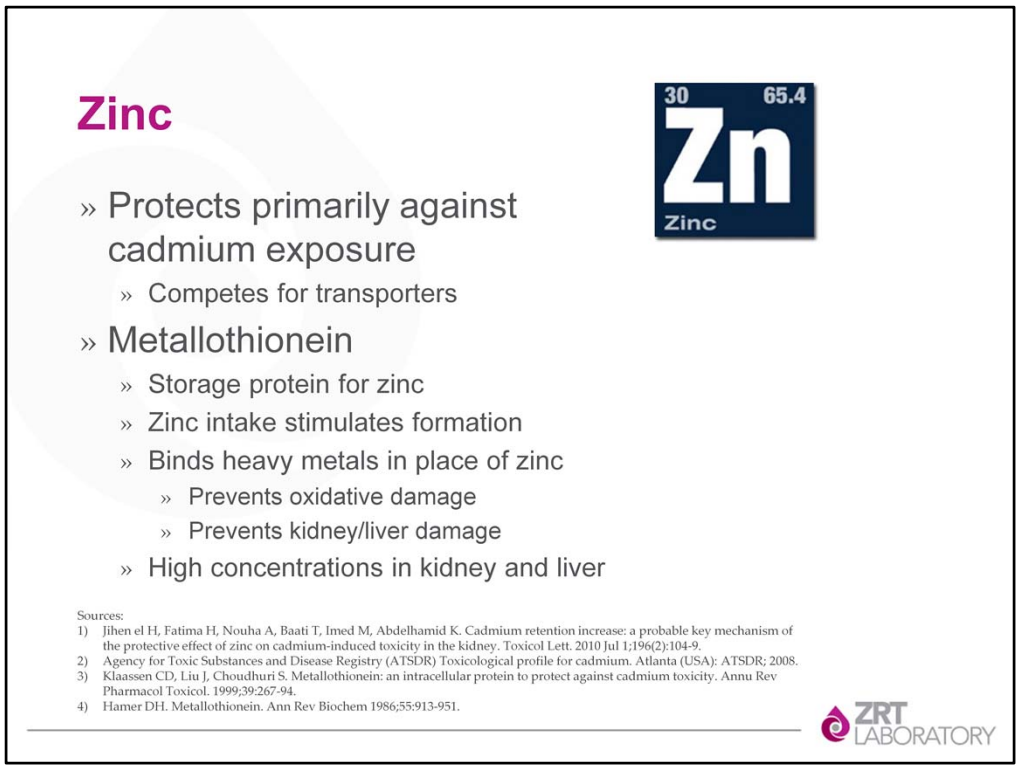
Selenium is one of the most important elements for protection against heavy metals. Selenium is an essential part of seleno-protiens, many of which are important antioxidants. Glutathione peroxidase is undoubtedly the most important.

Heavy metals have a high affinity for selenium, binding tightly to the essential element and preventing it from being incorporated into seleno-protiens or by inactivating them. Mercury itself has a binding affinity with selenium that is 1 million times higher than that of sulfur. A diet rich in selenium will help provide an abundance of selenium that will reduce or prevent oxidative damage caused by heavy metals, and by binding directly to heavy metals, preventing them from causing damage.

Selenium sufficiency has also been shown to decrease lead absorption, as it is believed that selenium will bind to lead in the gut and prevent it from being absorbed.

It is important to note that excessive selenium consumption can be dangerous, making it important to determine intake prior to supplementation.

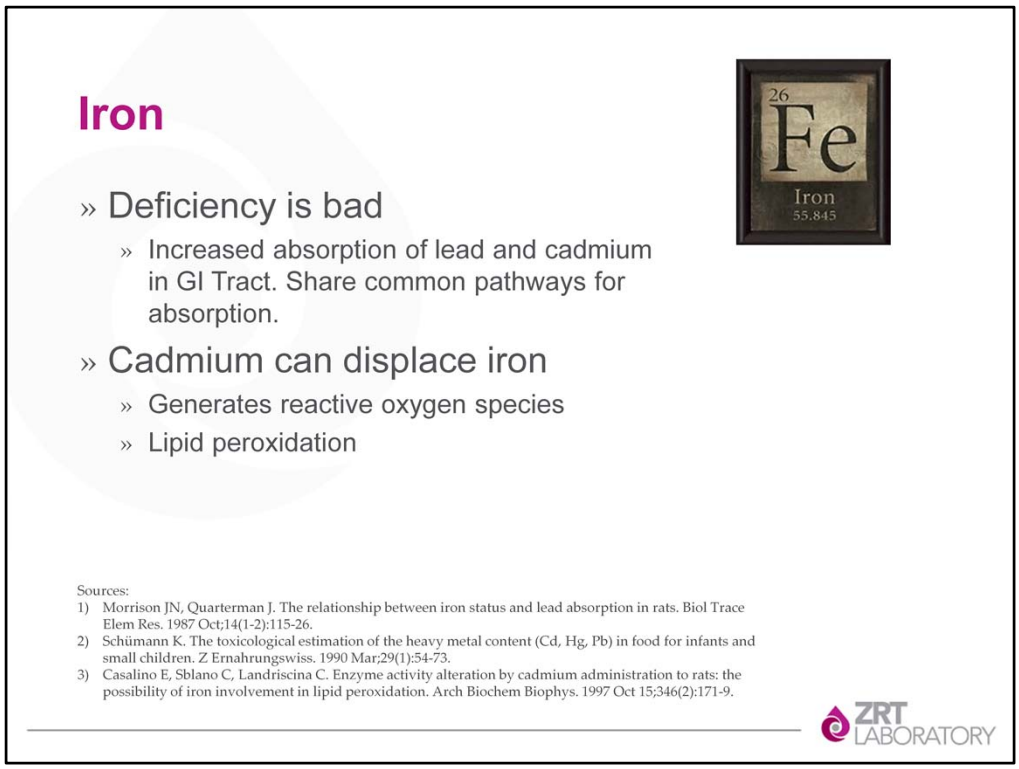
An interesting fact to share is that arsenic was once used to reduce selenium toxicity in cattle due to the elements interactions with each other. This is obviously not advised for humans.



Zinc is another important essential element for protection against heavy metals.

Zinc and cadmium share many properties and are often found connected to each other in nature. Proper zinc nutrition has been shown to decrease the toxicity of cadmium by competing for transporters within the body and preventing cadmium uptake.

Zinc intake also stimulates the production of metallothionein, a protein capable of storing zinc and binding heavy metals. There are 7 potential storage positions for zinc, which can be replaced by mercury and cadmium which have higher affinities for the cysteine residues than zinc. This locks up the heavy metals and prevents oxidative damage. Metallothionein levels are highest in the kidney and liver, which is why these two organs generally have the highest heavy metal burden.



Iron is important primarily in the prevention of cadmium uptake in the gastrointestinal tract. It has been shown that an iron deficiency, along with zinc and calcium, will increase cadmium absorption. This is believed to be one of the reasons why vegetarians, vegans, and women in general have higher body burdens of cadmium, as they may be more susceptible to iron deficiency.

When cadmium displaces iron, free iron may generate reactive oxygen species due to Fenton chemistry, which can result in lipid peroxidation.

ZRT Laboratory Heavy Metal Testing

- Element analysis by ICP-DRC-MS
  - Lowest Detection Limits
  - Highly Sensitive/Specific
- Excellent Proficiency Testing Performance
  - CAP, CDC, Interlaboratory
- Reference Ranges Established in House
  - Results compared to testing population

ZRT Reference Range	Zn (mg/L)	Cu (mg/L)	Mg (mg/L)	Se (µg/L)	Cd (µg/L)	Pb (µg/dL)	Hg (µg/L)	Zn:Cu Ratio
10-90%	6.35-9.35	0.79-1.14	36-57	170-318	<1.03	<2.50	<5.37	6.6-10.2

ZRT LABORATORY

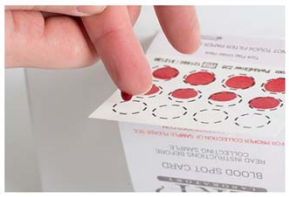
Now that we have talked about the big 4 heavy metals, what results from each sample type indicate, and essential protective elements, I would like to talk about the great testing options that we offer for toxic and essential elements at ZRT Laboratory.

Our element analysis is completed using a state of the art Perkin Elmer NexION 300D Inductively Coupled Plasma Mass Spectrometer with a dynamic reaction cell. This is abbreviated as I C P-D R C-M S. The use of a ICP-DRC-MS allows us to reach incredibly low limits of quantification, providing meaningful and quantitative low level results. Many other laboratories have high limits of quantification or only provide qualitative results for their elemental analysis, which is only useful for very high levels of exposure.

ZRT Laboratory participates in CAP and CDC proficiency testing programs for elements testing, assuring that results are accurate, precise, and comparable to liquid urine and whole blood.

Like all laboratories should do, we establish our reference ranges in house, which match well with established ranges provided by the NHANES US population surveys for urine and whole blood. It is important to know where an individual stands in reference to the population.

ZRT Laboratory Heavy Metal Testing Cont.



- Important to test Urine & Whole Blood Together!
- Testing with meaning!
- Dried Urine Test
  - **Arsenic**, **Cadmium**, **Mercury**, Selenium, Iodine, Bromine + Creatinine Correction
- Blood Spot Test
  - **Lead**, **Cadmium**, **Mercury**, Selenium, Zinc, Copper, Magnesium + Zinc/Copper Ratio

	Urine	Whole Blood
Arsenic	Recent Exposure	Cleared Rapidly
Lead	Chelation Testing?	Relatively Recent Exposure
Mercury	Elemental/Inorganic	Organic
Cadmium	Long Term Exposure	Relatively Recent Exposure

ZRT LABORATORY

At ZRT Laboratory we provide element results with meaning! Other laboratories may offer larger panels that focus on testing as many elements as possible, but we decided to go a different route. The element testing we provide only tests the most useful sample type, and we do not test for things that we believe will mislead the patient.

For example, testing only urine or only whole blood will not provide the whole picture for the heavy metals mercury and cadmium, and may mislead the patient. Blood and urine result in many cases do not match up because they mean two completely different things.

Each report we include with analysis is customized for the individual testing, and helps explain results for the patient and provider.

Dried Urine/Blood Spot Advantage

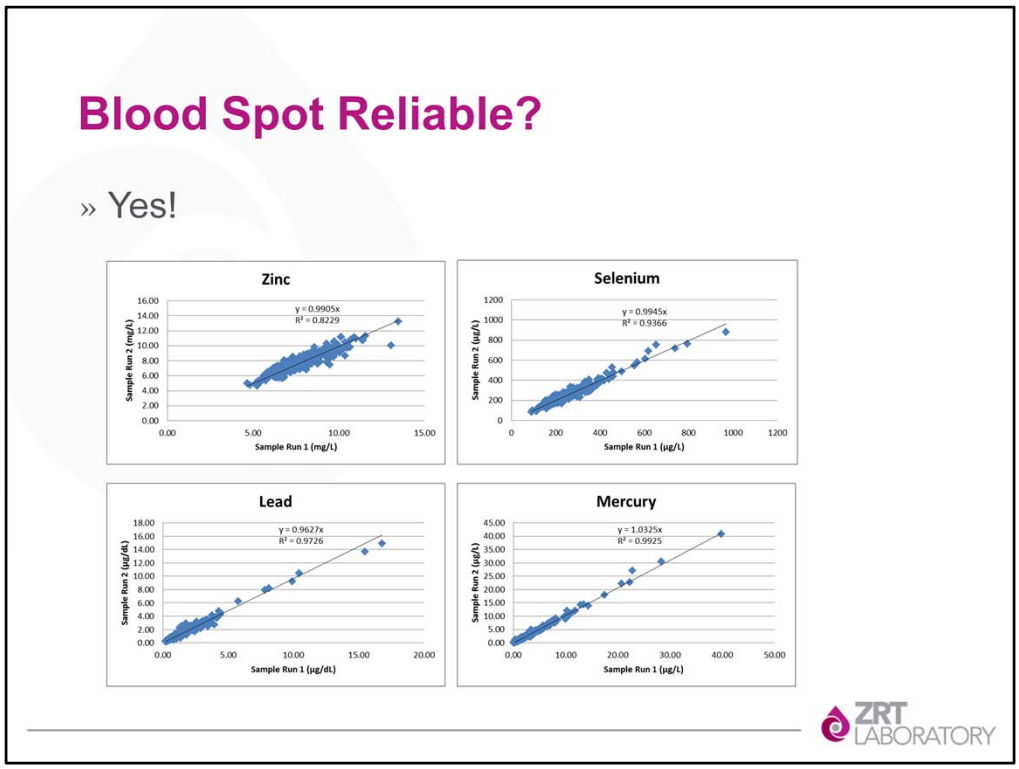
- Collection at Home
  - No 24hr jugs
  - No dangerous supplements
- Easy Domestic/International Shipping
  - Fits in an envelope without refrigeration
- Low Sample Volumes
  - Finger Prick > Venous Blood Draw
- Long Term Stability at Room Temperature

ZRT LABORATORY

There are many advantages to dried urine and blood spot testing over conventional liquid urine collection and venous blood draws.

Dried urine and blood spot can easily be collected at home, and do not require the use of the 24 hour collection jug, provocation agents, or loading doses. They are incredibly stable at room temperature, and do not require refrigeration or special handing at any point during shipment. This works especially well for international patients.

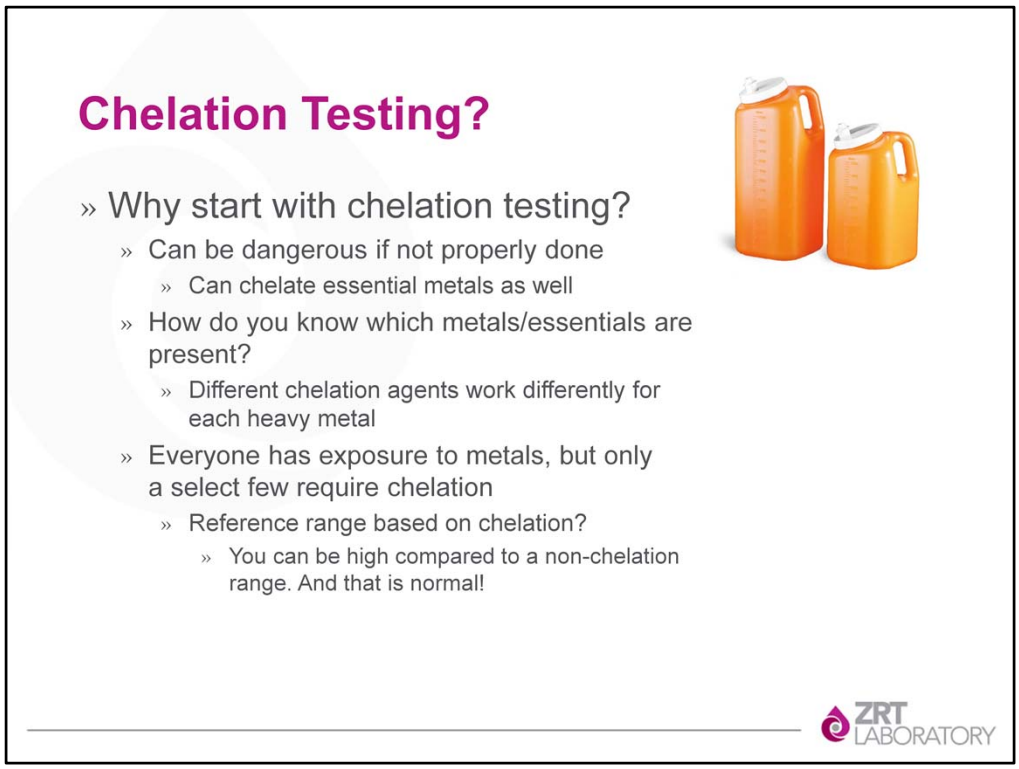
Our urine strips only require a couple mL of urine per strip, and our blood spot collection requires only a couple good drops of blood for analysis.



As far as I know, ZRT Laboratory is the only laboratory that has commercialized a multi-element blood spot assay. Many of you are probably wondering if blood spot is a reliable sample type for testing of essential and toxic metals.

The following 4 graphs are from the validation of our blood spot element assay.

We took 500 samples and ran them in duplicate, for a total of 1000 runs. We compared the first run to the second run to see if results could be replicated. As you can see, they match up very well!



What about chelation testing?

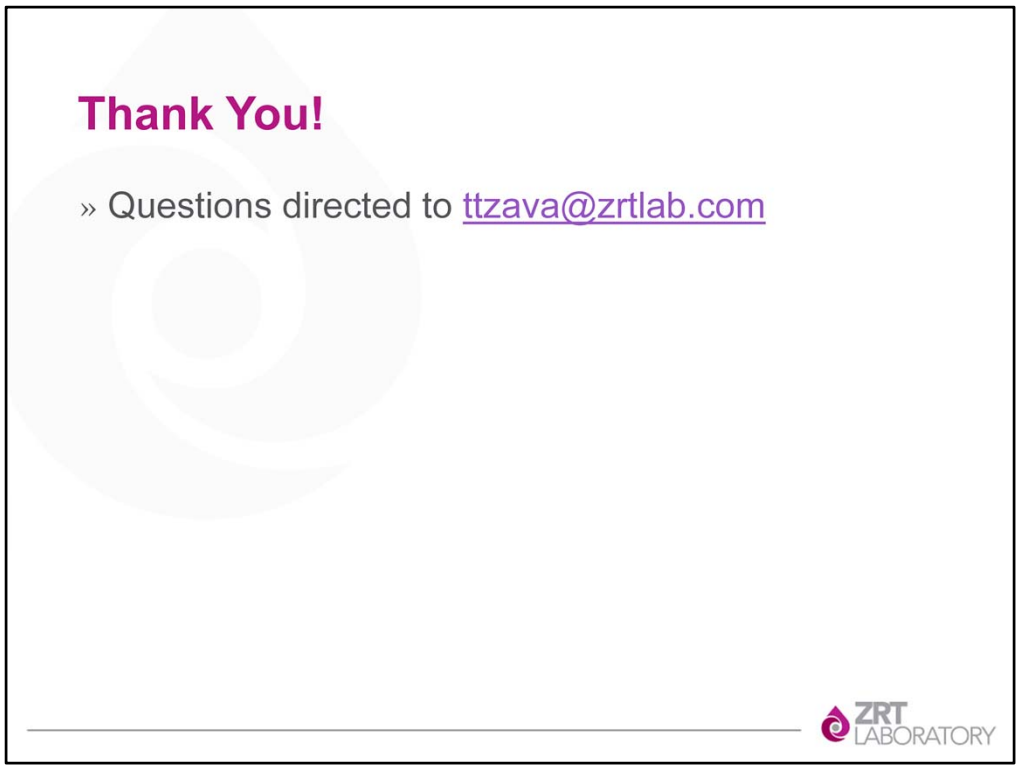
We are often asked if we can do chelation testing. The answer is yes and no.

Chelation testing is really only done using urine, so blood spot is not an appropriate sample type. Our urine collection consists of a morning and night spot sample collection, and does not involve collection into a container at any point. Chelation testing is typically done over 6, 12 or 24 hours because peak element excretion after taking a chelating agent will differ for each element and depends on the chelating agent. This makes it difficult to do chelation testing with spot samples, as you may miss the peak of excretion.

If you would like to do chelation testing with us, you will simply need to find a suitable urine collection device such as a 24 hour collection jug, collect all urine for the appropriate time period, then dip both strips into that collection. You will be responsible for doing all calculations after we send our results. It is important to understand that results will be compared to a non-chelation reference range, which may make it look like a patient has high toxic metal exposure when really results are normal for chelation testing. Everyone has metal exposure, but not everyone requires chelation treatment.

One thing that I have questioned recently is why chelation testing is done before a simple urine and blood screen for toxic and essential metals. Each chelating agent is different in what it best chelates, so why not figure out what to use before you blindly guessing? Also, chelation agents will remove essential elements as well, so it is important to do a screen

before chelation testing to determine if a patient is a good candidate for chelation. Depletion of essential elements may do more harm than good.



Thank you for your time and attention! If you have any questions after the lecture, feel free to send me an email if you think of a question at a later time. I will do my best to promptly answer it.

I will now turn the microphone over to Lissa who will explain about our new pricing for dried urine and blood spot element testing.

ZRT Lowers Elements Pricing

» **Dried Blood Spot Elements Profile:** Mercury, Cadmium, Lead, Zinc, Copper, Selenium, Magnesium

$99 BillMD

» **Dried Urine Elements Profile:** Iodine, Bromine, Selenium, Arsenic, Mercury, Cadmium, Creatinine

$99 BillMD

» **Comprehensive Elements Profile:** Blood Spot: Mercury, Cadmium, Lead, Zinc, Copper, Selenium, Magnesium

» Dried Urine: Iodine, Bromine, Selenium, Arsenic, Mercury, Cadmium, Creatinine

$159 BillMD

ZRT LABORATORY