MINISTRY OF HEALTH OF THE RUSSIAN FEDERATION
VOLGOGRAD STATE MEDICAL UNIVERSITY
DEPARTMENT OF PHARMACEUTICAL AND TOXICOLOGICAL CHEMISTRY

LECTURE 4

TOXICOLOGIKAL CHEMISTRY

Features of chemical-toxicological analysis for the content of arsenic compounds.

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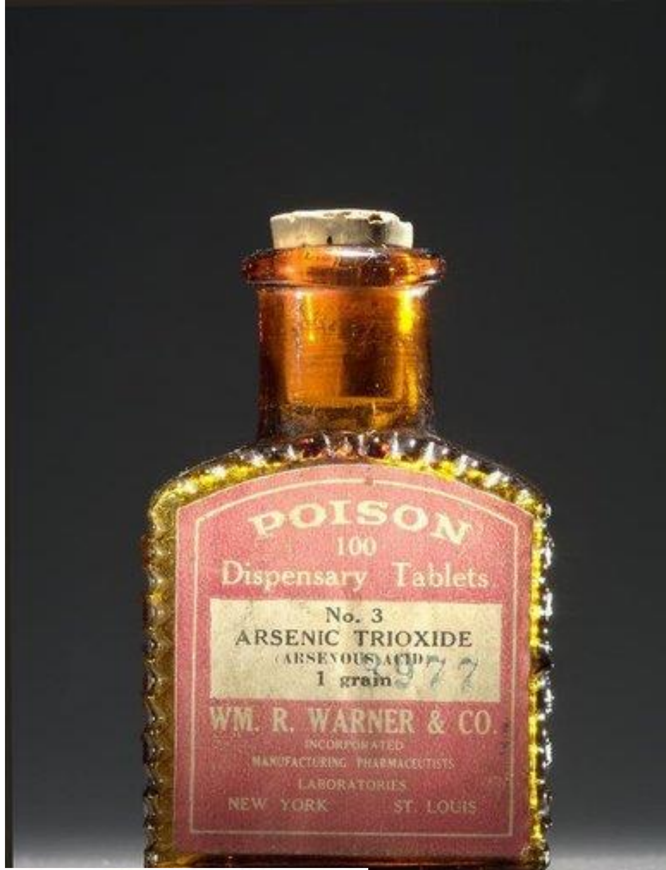
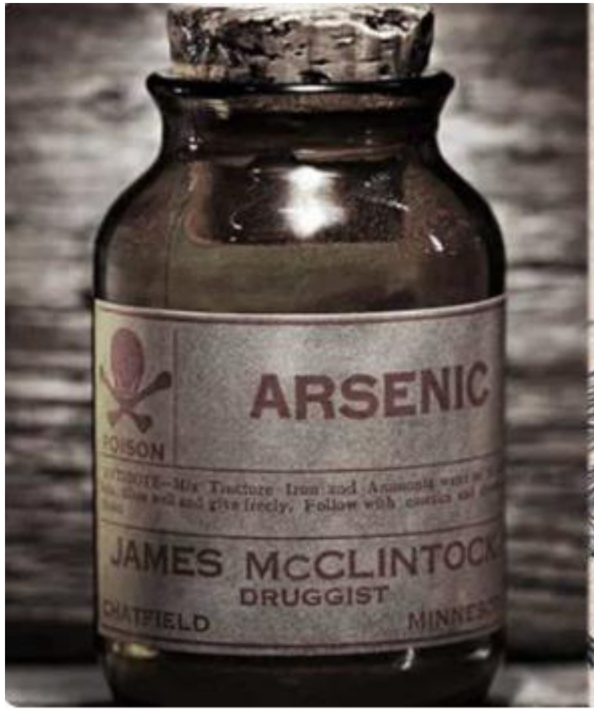
The metalloid arsenic is a natural environmental contaminant to which humans are routinely exposed in food, water, air, and soil.

Arsenic has a long history of use as a homicidal agent, but in the past 100 years arsenic, has been used as a pesticide, a chemotherapeutic agent and a constituent of consumer products.





Arsenic Was Once Marketed As A Beauty Product To Whiten Skin

Arsenic wasn't just known as a poison – it was also marketed as a way for women to beautify themselves. In the 19th century, women used arsenic powder to whiten their skin. It literally destroyed their red blood vessels, giving women the coveted pale, almost-dead look popularized in Victorian times. It could cause capillary damage in the skin, as well, which gave some a desirable "rosy-cheeked look."

According to an 1896 ad for Dr. Cambell's Safe Arsenic Complexion Wafers (pictured above), rubbing arsenic on your face would transform you into a "lovely creature" and an "object of worship." The wafers purported to remove pimples, freckles, and tans, and give skin radiant health. That is, if you survived the side effects.

L-O-V-E-L-Y!

She was certainly an exquisitely lovely creature. Nothing could have been added to enhance her beauty. She compelled **admiration**, and was an object of worship. This is the **universal result** of the use of

DR. CAMPBELL'S

SAFE ARSENIC COMPLEXION WAFERS

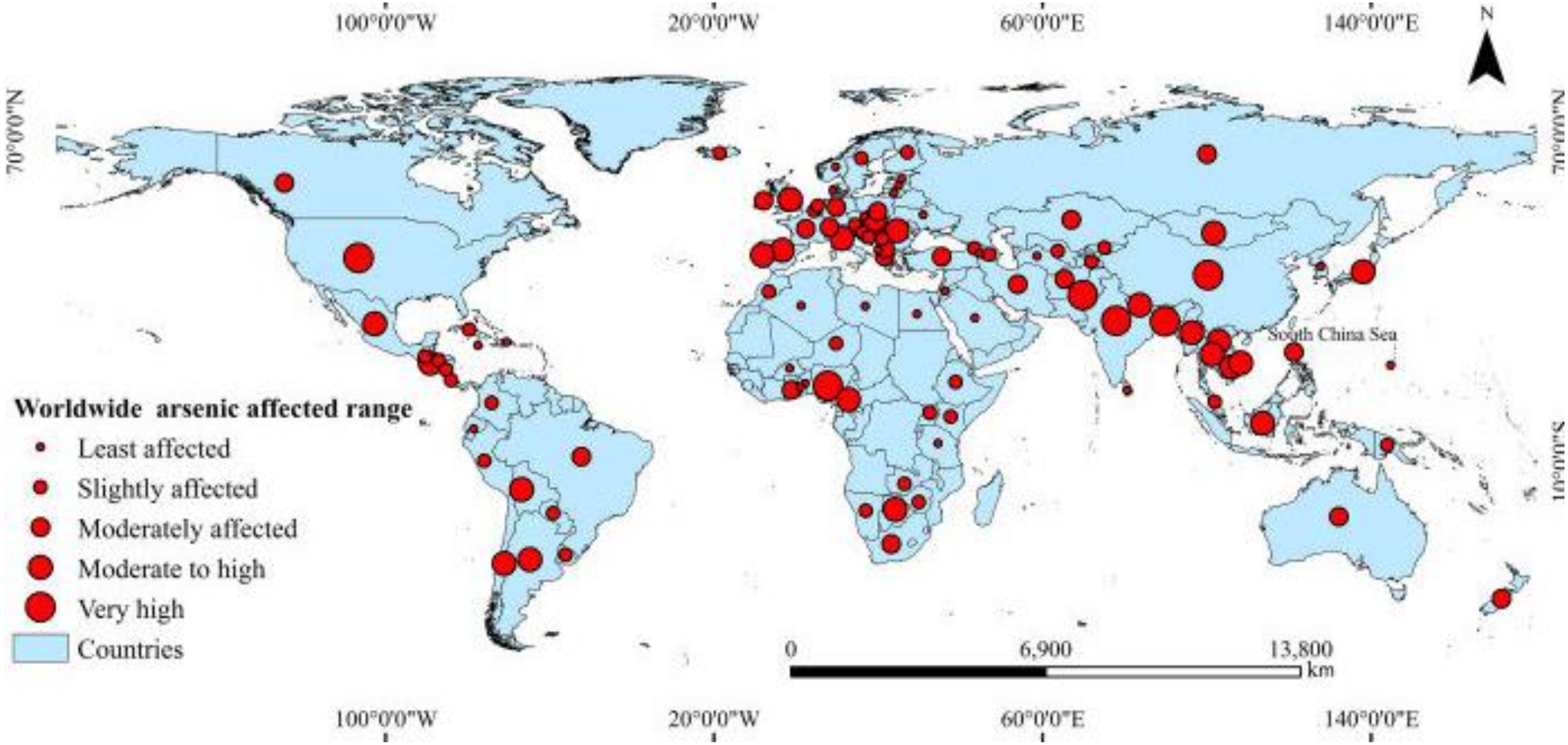
and **Fould's Medicated Arsenic Complexion Soap.** These two **World Famous Beautifiers** transform the most sallow skin into **radiant health**; remove **pimples**; clear the face of **freckles** and **tan**; give the complexion **an indescribable brilliancy**, and lend to every young lady **a charm of person** which makes her **ADORABLE.** Wafers by mail 50c. and $1.00 per Box. 6 large boxes $5.00. Soap by mail 50c. per cake.

Address H. B. Fould, 214 6th Ave., New York.

SOLD BY DRUGGISTS EVERYWHERE.

Photo: Nesster / Flickr / CC BY 2.0

In some areas of the world, high levels of arsenic are naturally present in drinking water and are a toxicological concern.



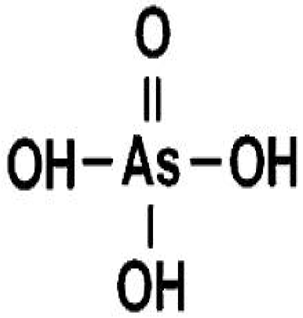
(www.who.int/news-room/fact-sheets/detail/arsenic; www.sos-arsenic.net/)).

There are several structural forms and oxidation states of arsenic because it forms alloys with metals and covalent bonds with hydrogen, oxygen, carbon, and other elements.
Environmentally relevant forms of arsenic are inorganic and organic existing in the trivalent or pentavalent state.

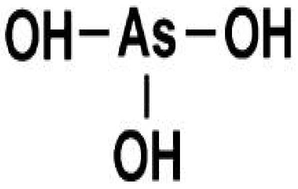
Metabolism of arsenic, catalyzed by arsenic (+3 oxidation state) methyltransferase, is a sequential process of reduction from pentavalency to trivalency followed by oxidative methylation back to pentavalency.
Trivalent arsenic is generally more toxicologically potent than pentavalent arsenic.

Arsenic Compounds of Environmental and Human Relevance

Trivalent oxidation state	Pentavalent oxidation state
Arsenite	Arsenate
Arsenic trioxide	Arsenic pentoxide
Monomethylarsonous acid	Monomethylarsonic acid
Dimethylarsinous acid	Dimethylarsinic acid
	Trimethylarsine oxide
	Arsanilic acid
	Arsenobetaine


$\mathrm{OH-\underset{\underset{OH}{|}}{\overset{\overset{O}{\|}}{As}}-OH}$

Arsenic Acid, Arsenate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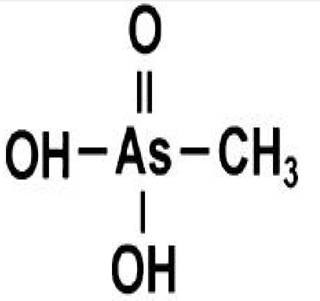
Arsenious Acid, Arsenite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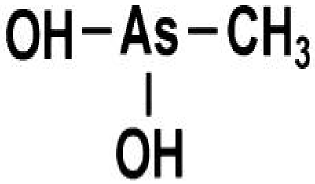
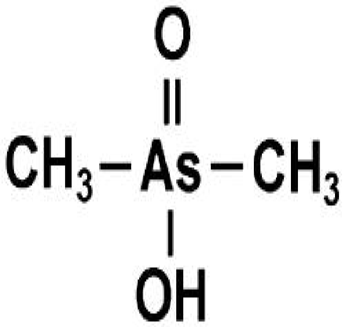
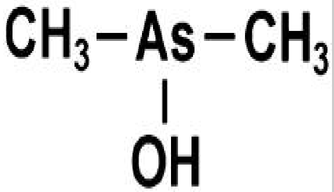
Monomethylarsonic Acid


$\mathrm{OH-\underset{\underset{OH}{|}}{As}-CH_3}$

Monomethylarsonous Acid

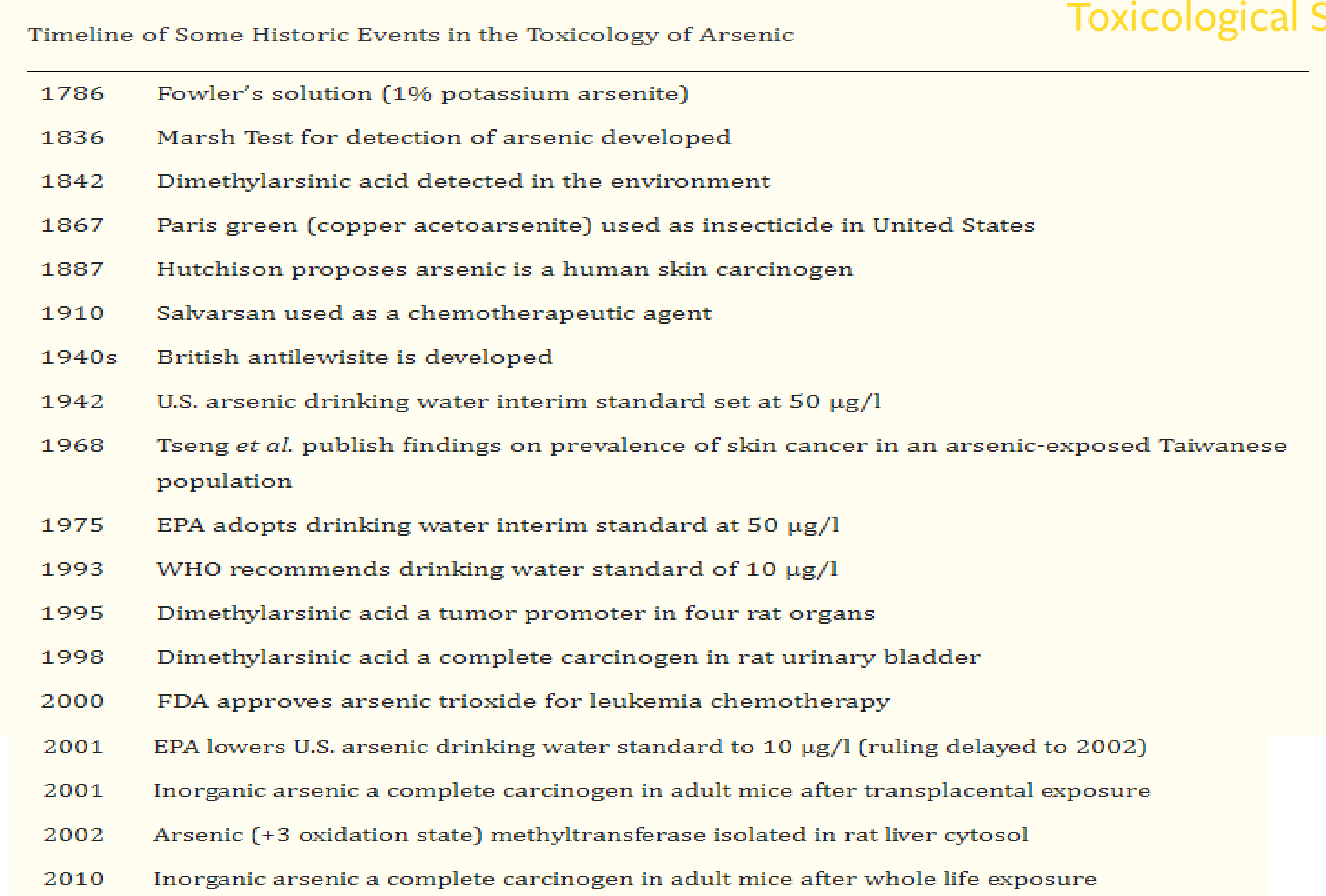

$\mathrm{CH_3-\underset{\underset{OH}{|}}{\overset{\overset{O}{\|}}{As}}-CH_3}$

Dimethylarsinic Acid


$\mathrm{CH_3-\underset{\underset{OH}{|}}{As}-CH_3}$

Dimethylarsinous Acid

Toxicological Sciences

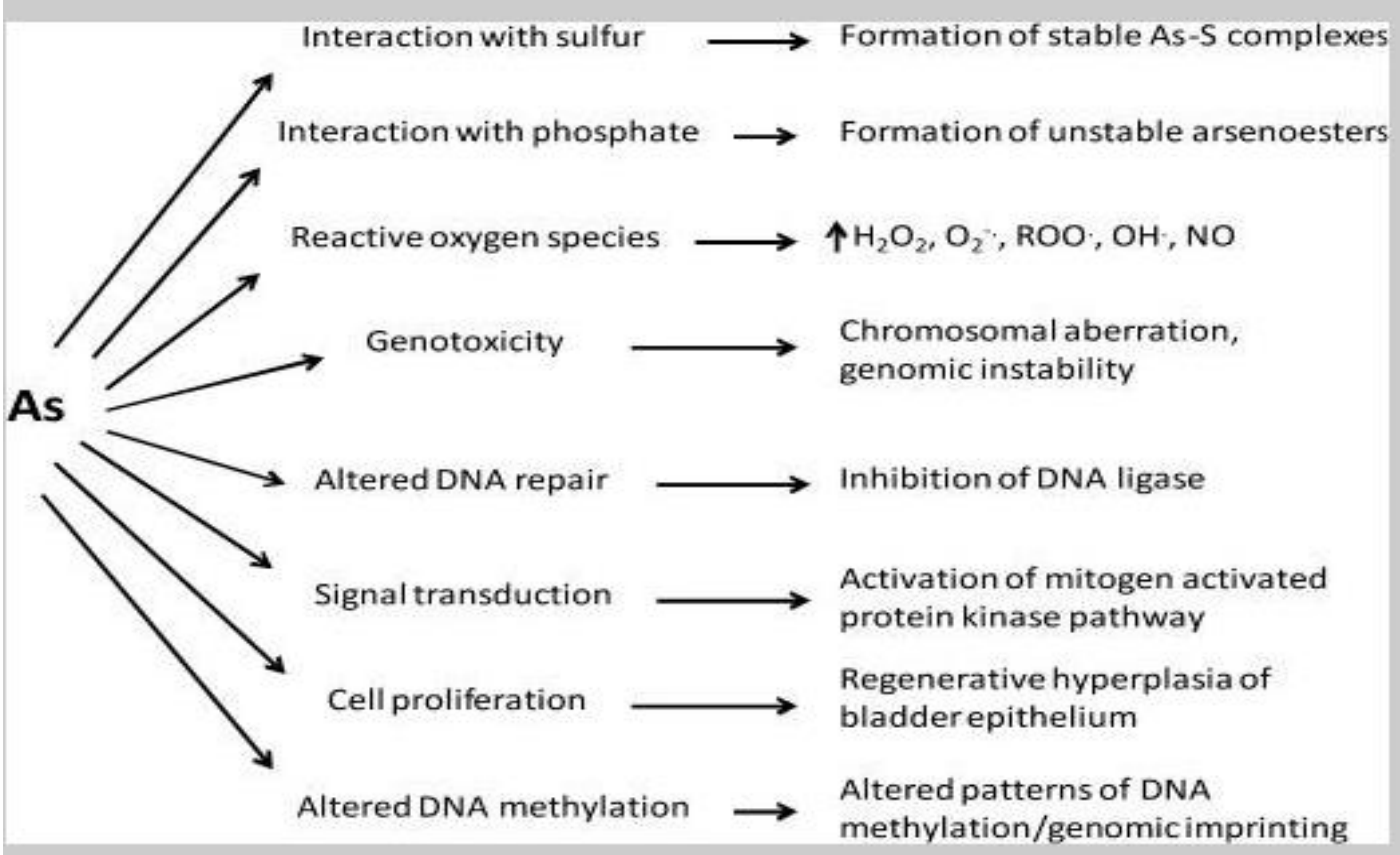


Timeline of Some Historic Events in the Toxicology of Arsenic

1786	Fowler's solution (1% potassium arsenite)
1836	Marsh Test for detection of arsenic developed
1842	Dimethylarsinic acid detected in the environment
1867	Paris green (copper acetoarsenite) used as insecticide in United States
1887	Hutchison proposes arsenic is a human skin carcinogen
1910	Salvarsan used as a chemotherapeutic agent
1940s	British antilewisite is developed
1942	U.S. arsenic drinking water interim standard set at 50 μg/l
1968	Tseng *et al.* publish findings on prevalence of skin cancer in an arsenic-exposed Taiwanese population
1975	EPA adopts drinking water interim standard at 50 μg/l
1993	WHO recommends drinking water standard of 10 μg/l
1995	Dimethylarsinic acid a tumor promoter in four rat organs
1998	Dimethylarsinic acid a complete carcinogen in rat urinary bladder
2000	FDA approves arsenic trioxide for leukemia chemotherapy
2001	EPA lowers U.S. arsenic drinking water standard to 10 μg/l (ruling delayed to 2002)
2001	Inorganic arsenic a complete carcinogen in adult mice after transplacental exposure
2002	Arsenic (+3 oxidation state) methyltransferase isolated in rat liver cytosol
2010	Inorganic arsenic a complete carcinogen in adult mice after whole life exposure

Acute effects of arsenic range from gastrointestinal distress to death. Depending on the dose, chronic arsenic exposure may affect several major organ systems. A major concern of ingested arsenic is cancer, primarily of skin, bladder, and lung.

The mode of action of arsenic for its disease end points is currently under study. Two key areas are the interaction of trivalent arsenicals with sulfur in proteins and the ability of arsenic to generate oxidative stress.



As

Interaction with sulfur → Formation of stable As-S complexes

Interaction with phosphate → Formation of unstable arsenoesters

Reactive oxygen species → ↑H_2O_2, $O_2^{-\cdot}$, ROO·, OH·, NO

Genotoxicity → Chromosomal aberration, genomic instability

Altered DNA repair → Inhibition of DNA ligase

Signal transduction → Activation of mitogen activated protein kinase pathway

Cell proliferation → Regenerative hyperplasia of bladder epithelium

Altered DNA methylation → Altered patterns of DNA methylation/genomic imprinting

The main manifestations of excess arsenic:

- irritability, headache;
- liver dysfunction, development of fatty hepatitis;
- skin allergic reactions, eczema, dermatitis, itching, ulcers, skin depigmentation, palmoplantar hyperkeratosis, conjunctivitis;
- damage to the respiratory system (fibrosis, allergies, perforation of the nasal septum, tumor);
- vascular damage;
- nephropathy;
- increased risk of developing skin cancer, liver, lungs;

The toxic dose of arsenic for humans is 5-50 mg,
lethal dose of As_2O_3*- 50-340 mg.*

- **Symptoms of poisoning by compounds arsenic** develop in 1-2 hours. Marked disturbances in the gastrointestinal tract: smell of garlic from the mouth, nausea, vomiting, colicky abdominal pain, dry skin due to dehydration. Appear tachycardia, decreased blood pressure for acute intoxication intravascular hemolysis, acute renal, hepatic failure, cardiogenic shock; hoarseness voices, protein and blood in the urine, oliguria, then anuria. Convulsions, suffocation, loss of consciousness may occur, jaundice up to the development of toxic hepatitis. Poisoning with arsine [arsenic hydride, arsenous hydrogen (AsH3)] leads to hemolysis and methemoglobinemia, coma quickly develops.

Objects of study

1. **body of the living** take:

-blood, urine , hair, Nails, vomiting

2 **forensic expertise (dead):**

- *1/3 liver, 1 kidney, stomach with contents, blood, urine, ¼* of the lung

Chronic poisoning with arsenic compounds - hair, nails, flat bones.

Isolation method is mineralization

Mineralization methods were discussed in the last lecture!

Identification

To detect arsenic in mineralizate use:

1. **Atomic absorption spectrometry.**

Detection is carried out according to the characteristic of arsenic resonant transition lines at wavelength 193.7nm.

Evaluation of the method. Detection limit is 2 µg arsenic in 1 ml of the test sample. https://www.jove.com/v/55953/determination-inorganic-arsenic-wide-range-food-matrices-using?language=Russian

2. Chemical methods:

- **Sanger-Black reaction**,
- reaction with a solution of silver diethyldithiocarbamate in pyridine
- **Marsh reaction**.

Sanger-Black reaction

based on the reduction of arsenic compounds to arsenic hydrogen, which is then filter paper reacts with mercury(II) chloride or bromide. The reaction is carried out in a specialdevice.

1 – flask;
2 – nozzle filled with cotton wool, soaked in acetate solution lead;
3 – paper soaked in solution mercury(II) chloride or bromide.

The reduction of arsenic compounds is carried out with hydrogen at the moment of its release, which is obtained by the interaction of a metal zinc with sulfuric acid:

$Zn + H_2SO_4 \rightarrow ZnSO_4 + 2H$

Zinc metal and sulfuric acid used to produce hydrogen, must not contain arsenic. Reaction between metal zinc and sulfuric acid proceeds slowly. To speed it up, so-called “cupped” zinc is used.(zinc, the surface of which is coated with copper sulfate).Hydrogen formed by the reaction of sulfuric acid andzinc, reduces arsenic compounds to AsH_3:

$AsO_2^- + 7H \rightarrow AsH_3 + H_2O$

$AsO_33^- + 9H \rightarrow AsH_3 + 3H_2O$

$AsO_43^- + 11H \rightarrow AsH_3 + 4H_2O$

The resulting arsenic hydrogen reacts with chloride or mercury(II) bromide, which is impregnated with filter paper. Atreaction produces a series of colored compounds that are located onpaper in the form of yellow or brown spots.

$AsH_3 + HgCl_2 \rightarrow AsH_2(HgCl) + HCl$

$AsH_3 + 2HgCl_2 \rightarrow AsH(HgCl)_2 + 2HCl$

$AsH_3 + 3HgCl_2 \rightarrow As(HgCl)_3 + 3HCl$

After treating the paper with a solution of potassium iodide all paper (except for a spot containing the indicated compounds arsenic) acquires a reddish color due to the transition of mercury chloride or bromide to iodide of this metal:

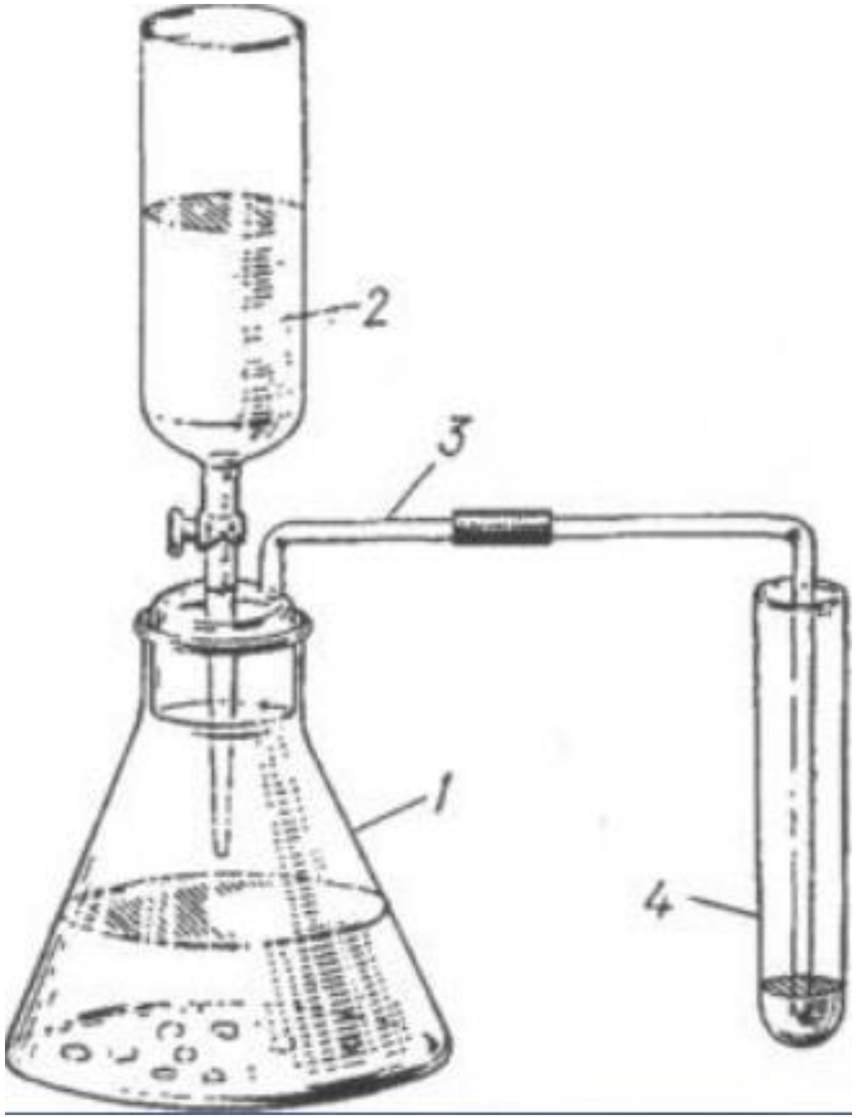
$HgCl_2 + 2KI \rightarrow HgI_2 + 2KCl$

The Sanger-Black reaction is interfered with by hydrogen sulfide, which can formed by the interaction of hydrogen with sulfuric acid:

$H_2SO_4 + 8H \rightarrow H_2S + 4H_2O$

Hydrogen sulfide released during interaction hydrogen with sulfuric acid, reacts on filter paper with mercury(II) chloride or bromide. As a result of this reaction black mercury sulfide is formed, which masks coloring of stains containing arsenic compounds. For tying hydrogen sulfide, use cotton wool soaked in acetate solutionlead:

$H_2S + Pb(OOCCH_3)_2 \rightarrow PbS\downarrow + 2CH_3COOH$



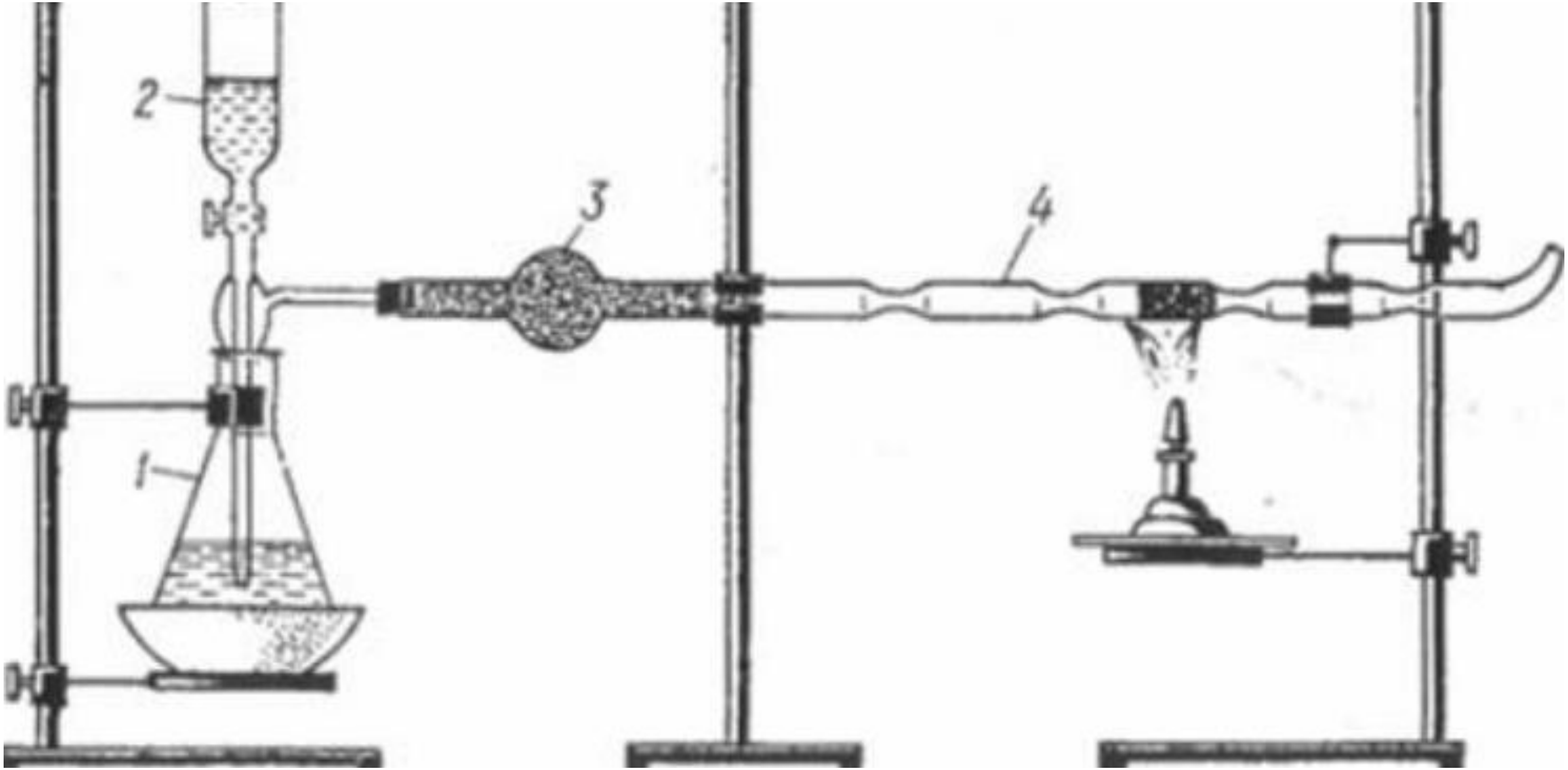
Reaction with silver diethyldithiocarbamate solutionin pyridine.Arsenic compoundsrestored toarsenous hydrogen, whichcollected in a test tube (receiver),containing freshly prepareddiethyldithiocarbamate solutionsilver in pyridine. Solutionsilver diethyldithiocarbamateshould not contain pyridinemoisture. If arsenic is present inmineralizate solutionsilver diethyldithiocarbamatetakes on a red-violet color https://en.wikipedia.org/wiki/Marsh_test

The Marsh reaction is based on the restoration of connections arsenic with hydrogen at the time of its release and subsequently thermal decomposition of the resulting arsenic hydrogen to elemental arsenic, which is then oxidizes to arsenous anhydride:

$AsO_2^- + 7H \rightarrow AsH_3 + 2H_2O$

$2AsH_3 \rightarrow 2As + 3H_2$

Arsenic formed during thermal decomposition arsenic hydrogen, is deposited on the walls of the recovery tube of the Marsh apparatus in the form of plaque (“arsenic mirrors"). Marsh's reaction is the most conclusive of all reactions recommended for the detection of arsenic in various objects. It not only allows you to detect small amounts of arsenic, but also distinguish it from antimony.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3
4
1

Determination of arsenic using the Marsh reaction performed in three stages.

1. Checking the reagents for the absence of arsenic
2. Determination of arsenic in the studied mineralizate
3. Authentication of plaque formed in recovery tube.

1. **Checking the reagents for the absence of arsenic** it is necessary to ensure that the ones used for this purpose, reagents (“cupped” zinc andsulfuric acid) do not contain arsenic. A blank experiment is carried out. Must not be signs of arsenic in Marsh's apparatus.!!!Air entering the Marsh apparatus through drip funnel may cause an explosion this apparatus when heated by reducingtube or when igniting the gases escaping from it

2. Determination of arsenic in mineralizate

1. Check for plaque in the recovery are a tube of the Marsh apparatus. The presence of plaque, its appearance and location in the recovery tube may indicate the presence of arsenic in the sample.
2. Ignite the hydrogen coming out of the apparatus tube Marsha. the flame takes on a bluish color. The smell of garlic.
3. Cold porcelain is added to the specified flame. Lids or porcelain plates on which appears brownish-grayish coating
4. Recovery tube of the Marsh apparatus carefully turn 180°, and then the end is immersed in a 5% solution of silver nitrate, slightly alkalinized ammonia the solution will darken as a result of the formation metallic silver:

$\mathrm{AsH3 + 3AgNO3 \rightarrow AsAg3 + 3HNO3}$

$\mathrm{AsAg3 + 3AgNO3 \rightarrow AsAg3 \bullet 3AgNO3}$

$\mathrm{AsAg3 \bullet 3AgNO3 + 3H2O \rightarrow 6Ag\downarrow + H3AsO3 + 3HNO3}$

Plaque researchArsenic deposits are distinguished from deposits of other substances(antimony, selenium, sulfur, coal) by color and their location inrecovery tube. Arsenic deposits have a brownish-gray color with a metallic sheen, antimony deposits are matte black, selenium deposits are gray, and sulfur deposits are yellowish orslightly brown.If the conditions for destruction of biologicalmaterial in mineralizates may contain organic substances,which are deposited in the recovery tube in the formblack plaque (coal).Arsenic deposits are deposited in the constricted partrecovery tube immediately behind the place of its heating, andantimony deposits form on both sides of the heating siterecovery tube. This is explained by the fact that when heated, antimony hydrogen SbH3 decomposes more easily thanarsenic hydrogen. In addition, antimony is less volatile thanarsenic.

For further study of plaque formed in recovery tube, it is disconnected from the Marsh apparatus and perform a series of experiments. Recovery tube in the area. The plaque location is heated. In this case, oxidation occurs substances deposited in the tube. Coal and sulfur deposits disappear from tubes, since their oxidation produces gaseous products (sulfur (IV) oxide or carbon (IV) oxide). Plaques arsenic and antimony are oxidized and deposited as oxides in cold areas of the recovery tube. Arsenic oxide has the shape of octahedrons, and antimony oxide is amorphous. Crystal formation, having the shape of octahedrons, is one of the most important evidence of arsenic in mineralizate.

1. When hydrogen sulfide is passed through the recovery tube,containing oxides of arsenic or antimony, yellow arsenic sulfide is formedcolor, and antimony sulfide - red or black. When in actionconcentrated hydrochloric acid, the color of arsenic sulfide does not change, but Antimony sulfide becomes discolored:

$$Sb2S3 + 6HCl \rightarrow 2SbCl3 + 3H2S.$$

2. Arsenic deposits that form in the recovery tube,dissolve in freshly prepared sodium hypochlorite solution:

$$2As + 5NaOCl + 3H2O \rightarrow 2H2AsO4 + 5NaCl$$

Antimony deposits do not dissolve in sodium hypochlorite.

3. microcrystalloscopic reactions.

When treating plaqueconcentrated nitric acid they dissolve to formarsenic and metaantimony acids and with

$$3As + 5HNO3 + 2H2O \rightarrow 3H3AsO4 + 5NO$$

$$3Sb + 5HNO3 + 2H2O \rightarrow 3H3SbO4 + 5NO$$

they are carefully evaporated to dryness. apply drop by drop 5 N. solution hydrochloric acid and a crystal of cesium chloride. In the presence of antimonyColorless crystals are formed in the form of Cs2SbCI5 polyhedra.Arsenic compounds with this reagent do not give crystals. If to the specified solutionadd a crystal of cesium chloride and a crystal of potassium iodide, then arsenic givesred-orange precipitate Cs2AsJ5

Advantages and Disadvantages of Arsenic Detection according to the Marsh method

The Marsh method has a number of advantages over other methods for detecting arsenic:

1) the ability to repeatedly check the availability or absence of arsenic in the test sample;
2) clarity and evidence of the research;
3) Marsh's method is the only acceptable one in as a method for detecting arsenic in the practice of forensic analysis. At the same time, detection of arsenic using the Marsh method requires a significant amount of time from an expert chemist.

Quantitative determination of arsenic

Atomic absorption spectrometry.

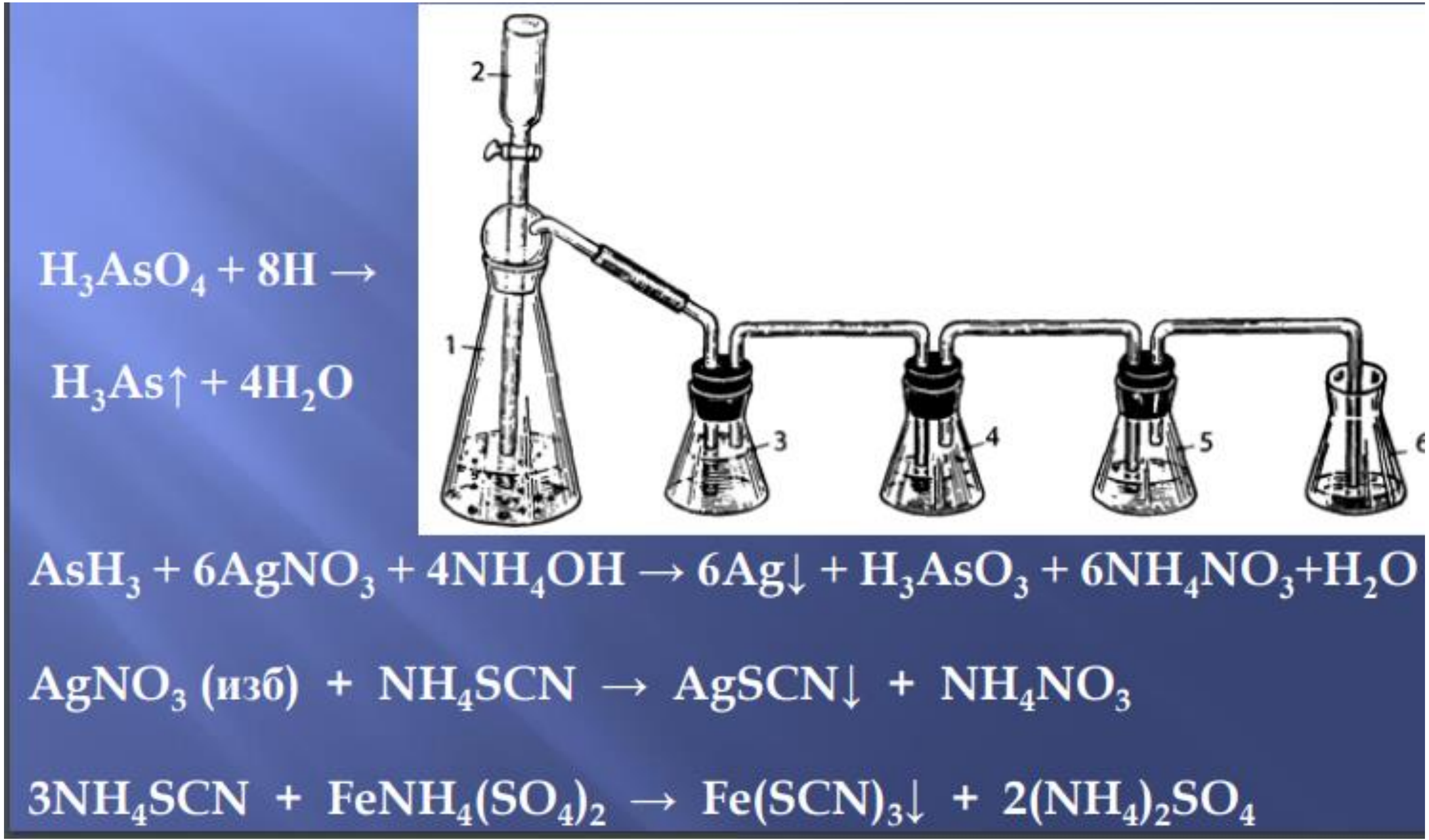
The determination is made by the amount of light absorption at wavelength 193.7 nm. Concentration calculations are carried out according to the calibration schedule or using the method additives

Quantitative determination of arsenic based on the reduction of arsenic in an acidic solution to arsenic hydrogen and its determination:

a) volumetric method;

b) colorimetric method according to SangerBlack.

a) Volumetric method (Volhard method) - based onreduction of arsenic acid in mineralizate to volatilearsenous hydrogen followed by its absorption with a titrated solution of silver nitrate.



$H_3AsO_4 + 8H \rightarrow$

$H_3As\uparrow + 4H_2O$

$AsH_3 + 6AgNO_3 + 4NH_4OH \rightarrow 6Ag\downarrow + H_3AsO_3 + 6NH_4NO_3 + H_2O$

$AgNO_3$ (изб) $+ NH_4SCN \rightarrow AgSCN\downarrow + NH_4NO_3$

$3NH_4SCN + FeNH_4(SO_4)_2 \rightarrow Fe(SCN)_3\downarrow + 2(NH_4)_2SO_4$

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3179678/

https://youtu.be/T6YSYOz-xyY?si=UT4sar92knjANnbG

https://www.jove.com/v/55953/determination-inorganic-arsenic-wide-range-food-matrices-using?language=Russian

https://www.youtube.com/watch?v=gtIbe2aD8a4

https://www.youtube.com/watch?v=P6q2viChrfM

POISON CHEMISTRY - WHITE ARSENIC

Arsenic is a notorious poison; colourless, odourless white arsenic was a popular choice for poisoners, and was commonly known as 'The King of Poisons'.

HISTORY

White arsenic has been known for centuries. In Ancient Rome, Nero's supposed use of it to poison his brother & become emperor is one of the first documented cases.

In the 17th & 18th centuries, white arsenic's use as a poison was widespread, and earned it the nickname 'inheritance powder'. However, its usage as a poison rapidly declined after the development of chemical tests.

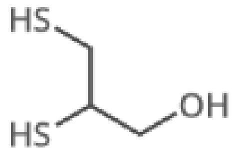
Around 50,000 tonnes of arsenic trioxide are still produced annually, and used as a precursor to a range of compounds. It's also been used as a treatment for some leukaemias.

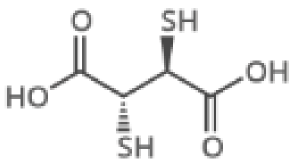
As_2O_3

ARSENIC (III) OXIDE

MEDIAN LETHAL DOSE: 15.1mg/kg

TREATMENT


DIMERCAPROL


DIMERCAPTOSUCCINIC ACID

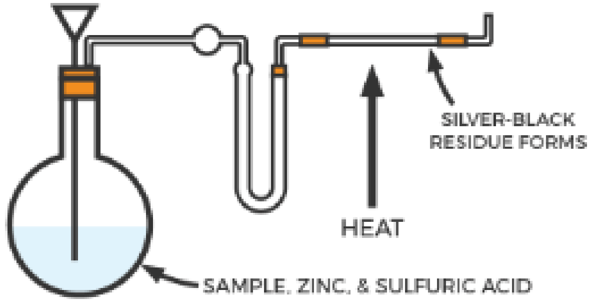
Chelating agents, such as the above compounds, bind the arsenic ions and prevent them from inhibiting enzymes. However, chelation therapy itself can have side effects. Dimercaprol has been largely superseded by 2,3-dimercapto-1-propanesulfonic acid.

EFFECTS

- HEADACHE
- DISCOLOURATION OF NAILS
- VOMITING & METALLIC TASTE
- BREATH SMELLS OF GARLIC
- STOMACH PAIN & DIARRHOEA
- HAIR LOSS
- CONVULSIONS
- CONVULSIONS, COMA & DEATH

Symptoms usually appear around 30 minutes after ingestion. Arsenic interferes with cell enzymes, respiration and mitosis. The skin, lungs, kidneys and liver are the major organs affected, with death occuring either from circulatory inefficiency, or liver or kidney failure.

DETECTION



The Marsh Test involves reaction of a sample with zinc and acid. If arsenic is present, it is converted to arsine gas. Heating arsine decomposes it; a silver-black deposit of arsenic is formed on cooling. Modern spectroscopic methods are now used instead of this test.