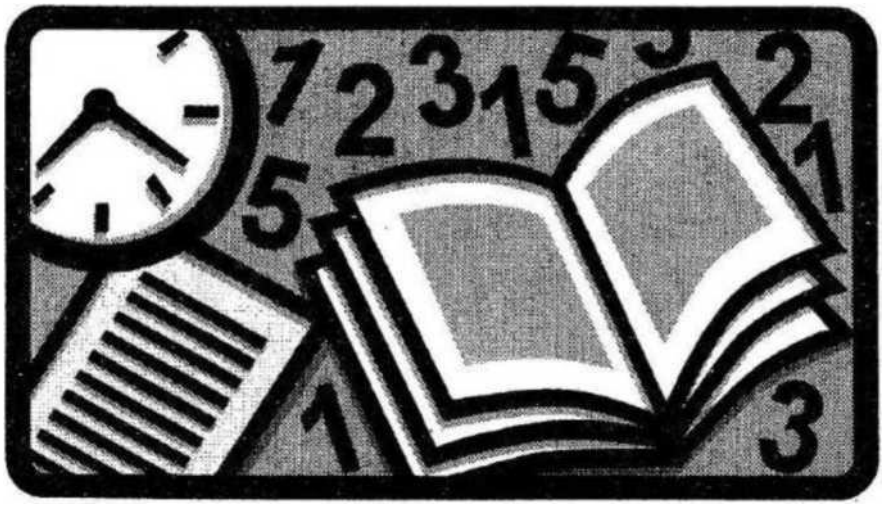
WORKING JOURNAL

IN TOXICOLOGICAL CHEMISTRY

STUDENT(S) OF THE GROUP COURSE



WORK RULES AND SAFETY PRECAUTIONS IN THE LABORATORY OF TOXICOLOGICAL AND FORENSIC CHEMISTRY

GENERAL SAFETY RULES

1. All work in the laboratory must be carried out in special clothing: gowns, caps and headscarves.
2. Work carefully, taking care not to introduce contamination into the reagents used in chemical toxicological analysis.
3. Based on the specifics of toxicological chemistry and the value of the reagents used, strive to study qualitative reactions with the smallest possible quantities and volumes of their solutions, using mainly slides and watch glasses, porcelain plates, cups and crucibles for reactions.
4. When performing tests in test tubes, do not heat their contents on an open flame of a gas burner, as this may cause hot liquid to escape with subsequent damage to the eyes, face and hands. Heating of test tubes with solutions should be performed in a water bath, directing the opening of the tube away from yourself and other workers, constantly stirring the contents of the tube by gentle shaking.
5. All work with substances that produce gases and odourous compounds that are hazardous to the body must be carried out under a fume hood. It is strictly forbidden to work with these substances in the workplace!
6. To prevent damage to the sewage system in the laboratory, solutions of acids, alkalis, and other aggressive substances must be poured into specially designed containers. Solutions of iodides, argentium and mercury compounds should be poured into separate containers.
7. It is forbidden to have personal belongings at the workplace, except for study guides and work logs.
8. Keep your hands clean during and after the work is completed. It is recommended to wash them first with water and then with soap and water.
9. After work, wash and put back the used dishes and reagents, and switch off the devices!

RULES FOR WORKING WITH TOXIC SUBSTANCES AND BIOLOGICAL MATERIAL

1. When working with poisonous, potent substances and biological material, personal precautions and safety measures must be strictly observed:
   a) do not touch poison and biological material with unprotected hands;
   b) not to store or take food and water in places where toxic substances and biological material are stored and handled.
2. When working with concentrated acids and alkalis, be careful not to get them on your clothes or skin.
3. When diluting concentrated sulphuric acid, be careful to add the acid to the water and not vice versa.

4. Alkalis that are in a solid state (potassium hydroxide, sodium hydroxide) must be collected from the container with tweezers or a spatula, and the grinding of the pieces must be carried out with special protective goggles, as small pieces that can fly off are very dangerous for the eyes and hair.

5. Dilute solutions of acids and alkalis are also hazardous to the eyes and skin, so care must be taken when handling them.

6. With biological material, it is necessary:

a) wear rubber gloves;

b) disinfect all potentially contaminated materials used in laboratory tests. Immediately after completion of work, wipe the premises and equipment with a 3% solution of chloramine B or a 3% clarified solution of bleach;

c) Boil protective clothing (gowns, scarves, hats, cotton gauze masks, gloves) in a 2% soda solution or any detergent for 30 minutes, or place in a 3% chloramine B solution for 2 hours at a rate of 5 l/kg. Autoclaving with a steam-air mixture at a temperature of 80-90° for 45 minutes is preferable.

RULES FOR WORKING WITH COMBUSTIBLE AND FLAMMABLE SUBSTANCES

1. Flammable substances (organic solvents) must be heated without fire, in a preheated water or other bath.

2. Add flammable liquids to the mixture of reactants from a small container (test tubes, flasks).

3. Dismantling of devices containing combustible substances must be carried out after work has been completed with the heating devices switched off.

4. Do not store combustible and flammable substances near a fire or highly heated electrical appliances.

5. Certain gases (hydrogen, acetylene, carbon monoxide), alcohols, flammable hydrocarbons (benzene, hexane), acetone, diethyl ether and other substances may form explosive mixtures with air. Work with such substances must be carried out with exhaust ventilation switched on to prevent the formation of dangerous concentrations of vapours and gases in the room.

6. Do not discharge used flammable liquids into the sewer! They must be collected in special hermetically sealed containers, which are taken out of the laboratory at the end of the working day.

RULES FOR WORKING WITH ELECTRICAL APPLIANCES

1. To prevent accidents, the use of electrical equipment (photoelectrocolourimeters, spectrophotometers, gas chromatographs, etc.) is prohibited:

* use appliances with damaged wiring insulation;
* leave appliances switched on unattended;
* connect the equipment to a network whose voltage does not correspond to the voltage required for the operation of the devices;
* replace blown fuses with wire;
* work with ungrounded devices.

2. The floor area near electrical appliances should be covered with a rubber mat.
3. When working with electrical appliances, at least two people should be present in the room.

FIRE FIGHTING

1. In the event of a fire, you must:
* Immediately switch off electric heaters and ventilation;
* Remove all containers with flammable substances from the laboratory;
* call the fire brigade by phone and inform the work supervisor and the head of the department;
* apply the most effective means of extinguishing the fire in the given case.
2. The flame must be extinguished by the following means:
* in case of ignition of liquids miscible with water (alcohol, acetone) - any fire extinguishers, water flow, sand, asbestos or cloth blanket;
* when igniting liquids that do not mix with water (petrol, petroleum ether, etc.), use carbon dioxide and powder fire extinguishers, sand, blanket, do not use water;
* De-energise burning wires and live electrical appliances and extinguish them with carbon dioxide fire extinguishers;
* burning wooden parts - with all extinguishing agents;
* if the employee's clothing catches fire, cover the burning area with something such as a towel, bathrobe, blanket or thick cloth.

PROVIDING FIRST AID TO VICTIMS OF ACCIDENTS

1. In case of contact with concentrated sulphuric acid, gently wipe it off with a dry cloth or cotton swab, wash the affected area with water and sodium bicarbonate solution. Other strong acids shall be gently washed off with water and then with sodium bicarbonate solution.
2. Wash the diluted acids off the affected area quickly, then treat the skin or eyes with 1% sodium bicarbonate solution, and then again with water.
3. In case of contact with concentrated caustic alkalis, rinse the affected area with water and neutralise with dilute acetic or citric acid.
4. In case of contact with eyes or skin with dilute alkali solutions, rinse with water, 1% boric acid solution, and then with water again.
5. In case of contact with phenol, bromine and other irritating substances, the damaged area should be washed with an organic solvent (alcohol, ether).
6. In case of body burns, the affected area should be washed with 5-10% potassium permanganate solution and a tampon soaked in 5% tannin solution or a special ointment for burns should be applied.
7. Cut areas should be treated with a special alcohol iodine solution and bandaged. Do not wash the wound with water or remove clotted blood.
8. In all cases of poisoning, the victim must first be taken out or into fresh air and assisted until the arrival of a doctor: free the victim from constricting clothing, and cover him/her warmly if necessary.

9. In case of electric shock, it is necessary to: turn off the lever or remove the fuse, carry the victim away from the place of injury and lay him/her on a flat place, unfasten the belt, give him/her an ammonia solution, and ensure complete rest.
10. After providing first aid, the victim should be immediately referred to a medical facility.

SESSION NO.

Date.

TOPIC: *PREPARATION OF A CHEMICAL TOXICOLOGICAL STUDY PLAN. EXTERNAL INSPECTION AND PRELIMINARY TESTING OF THE OBJECT OF STUDY.*

1. External inspection:

- determine the consistency and morphological composition of the object;
- determine the presence of a preservative;
- identify the smell and colour of the object;
- note the presence of foreign inclusions.

Based on your research, fill in Table 1.

No. of the object	odour	colour	morphological composition of the object	presence of a preservative	foreign inclusions

2. Preliminary testing of the facility.

2.1. Determine the reaction of the object's environment.

Procedure: Mix a small amount of the object in a test tube with an equal amount of distilled water. A part of the aqueous extract is applied to a universal indicator paper using a glass rod. A drop of distilled water (control sample) is applied to the other paper.

Chemical and toxicological assessment of the results: an acidic environment of the object excludes the study for the presence of alkalis in the object, an alkaline environment - xylenes. A strongly acidic environment (pH 1-3) may be caused by the presence of mineral acids or a large amount of organic acids introduced from the outside; an alkaline environment - by the presence of caustic alkalis, ammonia, carbonates and silicates, salts of weak acids and strong bases (cyanide, nitrite, etc.).

2.2 Difference between caustic alkalis and carbonate alkalis.

Methodology: Place 1-2 ml of the aqueous extraction with alkaline medium in a test tube. Add 1-2 drops of an alcohol solution of phenolphthalein (1:1000) - a pink colour is formed, then add a solution of barium chloride and shake: the colour may disappear or remain.

Chemical and toxicological evaluation of the results: **if the** colour of the solution remains, the medium is caused by the presence of free hydroxyl groups, if it disappears and a white precipitate forms, it is caused by carbonates or silicates.

2.3 Testing for ammonia and hydrogen sulphide.

Procedure: the object of the alkaline reaction is placed in a test tube, closed with a cork stopper, to the lower surface of which 3 pieces of paper are attached: a *wet red litmus test, a piece of paper moistened with a solution of copper sulphate and a piece of paper moistened with a solution of plumbum acetate.*

During the test, you can observe changes in the colour of all three papers or only two (copper and litmus).

Chemical and toxicological assessment of the results: if the colour of all three papers changes, it can be concluded that the object contains ammonia and hydrogen sulphide formed during the decay of biological material. If the colour of the copper and lamine papers changes, it can be assumed that ammonia has been introduced into the object.

Based on your research, fill in Table 2.

_____SESSION NO.

Date.

SUBJECT: *A GROUP OF SUBSTANCES THAT IS ISOLATED FROM BIOLOGICAL MATERIAL BY MINERALISING THE BIOLOGICAL MATERIAL AND REMOVING OXIDANTS FROM THEMINERALISATE.*

1. Mineralisation of the biological material with a mixture of sulphuric and nitric acids. The crushed biological material is placed in a Kjeldahl flask. Then, for every 100 g of the sample, add 75 ml of a mixture consisting of 25 ml of concentrated sulfuric acid, 25 ml of concentrated nitric acid and 25 ml of water. After the foaming of the mixture stops, the flask is fixed on a tripod and heated over an asbestos mesh (at a distance of 1-2 cm on a bare fire) until the solid lumps of the object material are destroyed and a dark liquid is obtained. After that, the heating temperature is increased, and nitric acid (1:1) is added to the reaction flask in small portions until the contents become colourless or slightly yellowish. Then heat the mixture without adding nitric acid until thick white sulphur oxide vapour appears. If the liquid does not darken, the destruction is complete, otherwise, the destruction of the object continues. After the mineralisation is complete, the liquid is cooled, transferred to a beaker and checked for oxidants by mixing a drop of mineralisate with a drop of diphenylamine in concentrated sulphuric acid. A blue colouration indicates the presence of oxidants that destroy formalin. To do this, dilute the mineralisate with a small amount of water (10-15 ml), heat to a boil, and add 0.5-1.0 ml of formalin dropwise until the gas bubbles stop. If necessary, the oxidant destruction operation is repeated. After that, the mineralisate is diluted with water to 180-190 ml.

2. Mineralisation of biological material with a mixture of sulphuric, nitric and chloride acids. 100 g of the crushed organ is placed in a Kjeldahl flask, to which 25 ml of water, 25 ml of concentrated nitric acid, 25 ml of concentrated sulfuric acid and 25 ml of 42% hydrochloric acid solution are added in the specified sequence. Next, the destruction of organic matter is carried out as described above.

The mineralisation obtained by either method is left for 18-20 hours at room temperature. If a precipitate forms, it is filtered and analysed for plumbum and barium (if the precipitate is dirty green, it is additionally analysed for chromium).

The mineralisate (filtrate after separation of barium and plumbum sulphates) is diluted with water to 200 ml and analysed for manganese, chromium, argentum, copper, cadmium, bismuth, zinc, arsenic, stibium, and thallium.

3. Mineralisation of organic substances by fusion with an oxidising agent. Mix 1-2 g of the object with a mixture of carbonate and sodium nitrate (2:1) in an amount of 4-6 g, then moisten with water and dry by heating in a water bath. Melt 5-6 sodium nitrate in a crucible. Reducing the flame of the burner under the crucible, the mixture to be analysed is added in separate portions. After the first portion is burned, add the second, then the third, and so on until all portions of the mixture are burned. After the last portion of the mixture has been burned, the vessel in which it was contained is rinsed with 2-3 g of sodium carbonate, after which this portion is also burned. During the melting process, care is taken to ensure that the flame in the crucible does not ignite, and the burner flame is regulated. The alloy is cooled and treated with boiling water. The resulting solution is tested for "metal" poisons (arsenic, argentum, and some others, except for mercury).

4. Mineralisation of organic matter by dry ashing. The object is crushed, dried in a sand bath, and then the sample is charred under gentle heating. The residue in the crucible after charring is

moistened with a concentrated solution of ammonium nitrate or concentrated nitric acid, dried in a boiling water bath, and heated over a low flame burner (the bottom of the crucible should not touch the flame). If necessary (incomplete combustion of organic matter - the ash is black or grey), the contents of the crucible are moistened again with a concentrated solution of ammonium nitrate, dried and calcined. The contents of the crucible are cooled, treated with hydrochloric acid (for manganese) or nitrate acid (for copper), the solution is filtered, the filtrate is evaporated in a water bath to dryness, the residue is dissolved in 3-5 ml of water and analysed.

SESSION NO.

Date.

SUBJECT: *MATERIAL TESTING FOR BARIUM, PLUMBUM, CHROMIUM AND MANGANESE.*

No. p/n	Methods of chemical reactions	Observations.	Chemistry of reactions	Chemical and toxicological assessment of reactions
1.	**Sediment research Barium detection Recrystallisation from concentrated sulphuric acid** **Methodology.** Place a grain of sediment from the filter on a slide, air-dry it, then apply 2-3 drops of concentrated sulfuric acid to the residue and heat it over a bare fire until white vapour appears, avoiding spreading. The glass with the drop is left in the air for 10-20 minutes. After that, the crystals are examined under a microscope.			
2.	*Independent work* **Production of barium iodate** **Methodology.** A grain of sediment is taken with a platinum loop or needle and placed first on the reducing part of the burner flame, then in a drop of 5M hydrochloric acid solution, and then on a slide. The operation is repeated several times. After that, a crystal of potassium iodate is added to the drop and examined under a microscope.			
1.	**Detecting plumbum Reaction with dithizone** **Methodology.** The test solution is placed in a test tube, 1 ml of 10% hydroxylamine chloride solution (but not sulphate) is added, pH is adjusted to 7.5-8 by adding 10% ammonium hydroxide solution, 2 ml of chloroform and a few drops of 0.01% dithizone solution in chloroform, and the mixture is shaken vigorously. The extraction of dithizone is continued until			

	the chloroform layer ceases to change from green to red. Next, the extract is re-extracted with 1M nitric acid solution, the re-extract is divided into four parts, and confirmatory tests are performed.			
2.	**<u>Reaction with potassium iodide</u>** **Methods.** Add a few drops of 5% potassium iodide solution to 1/4 of the solution.			
3.	**Reaction with sulphuric acid** **Methods.** Add a few drops of 10% sulfuric acid solution to 1/4 of the solution.			
4.	**Reaction with potassium dichromate Methods.** Add a few drops of fresh hydrogen sulphide water to 1/4 of the solution.			
1.	**<u>Leachate research</u> Manganese detection Oxidation with potassium peroxide Methods.** Add 4 ml of water, 1 ml of saturated sodium dihydrogen phosphate solution, 0.2 g of potassium peroxide to 1 ml of mineralisate and heat in a boiling water bath for 20 minutes.			
2.	**Oxidation with ammonium persulfate.** **Methods.** Add 4 ml of water, 1 ml of saturated sodium dihydrogen phosphate to 1 ml of mineralisate and heat in a boiling water bath for 5-6 minutes. Add 1 drop of a 10% solution of argentium nitrate, 0.5 g of ammonium persulfate to the hot solution and heat again until the persulfate is completely decomposed.			
1.	**Chromium detection Reaction with diphenylcarbazide Methods.** Add 4 ml of water, a drop of 10% argentum nitrate solution, 0.5 g of ammonium persulfate to 1 ml of mineralisate and heat in a boiling water bath for 20 minutes. Then add 1 ml of saturated sodium dihydrogen phosphate solution, potassium hydroxide solution (dropwise) to pH 2.0 and 1 ml of 0.25%			

	diphenylcarbazide solution to the mixture.			
2.	**<u>Formation of suprachromic acids</u> Methodology.** Add 30% sodium or potassium hydroxide solution to pH 7 to 5 ml of mineralisate dropwise, then add another 1 ml of mineralisate, then a drop of 10% argentum nitrate solution, 0.5 g of ammonium persulfate and heat in a boiling water bath for 20 min. The test tube with the test solution is cooled for 10-15 min in an ice bath. Add 1 ml of saturated solution of pH 2.0 to the cooled liquid, add ethyl acetate, but in the latter case, the aqueous layer must be saturated with an organic solvent beforehand. Then, 2-3 drops of 25-28% hydrogen peroxide solution are added to the test mixture and the contents of the test tube are immediately shaken vigorously.			

SESSION NO.

Date.

TOPIC: *ANALYSIS OFMINERALISATE FOR ARGENTUM, CUPRUM, ZINC, BISMUTH, ARSENIC, STIBIUM. DESTRUCTION OF BIOLOGICAL MATERIAL AND DETECTION OF MERCURY IN THE DESTRUCTATE.*

No. p/n	**Methods of chemical reactions**	**Observations.**	**Chemistry of reactions**	**Chemical and toxicological assessment of reactions**
1.	*Independent work* **Detection of argentum Reaction with dithizone** **Methods.** Add 1 ml of 4M sulfuric acid and 3 ml of 0.01% dithizone solution in chloroform or carbon tetrachloride to 5 ml of mineralisate. The mixture is shaken.			
2.	**<u>Production of argentium chloride</u>**			

	Methodology. Add 0.5 g of sodium chloride to 100 ml of mineralisate, heat the mixture to about 80° and leave for two hours or until the next day until a precipitate forms (sensitivity of the reaction is -2.5x10-^{2} mg per 100 ml, the reaction is specific). The precipitate is filtered off, washed with 0.5M hydrochloric acid, and then dissolved in 0.5-4 ml or more of 8M ammonium hydroxide solution. The ammonia filtrate is examined.			
3.	**<u>Formation of argentium iodide</u>** **Methods.** Add a saturated solution of potassium iodide to 1/3 of the filtrate solution (if its volume is more than 2 ml).			
1.	**Detection of copper** **Preliminary reaction with plumbum diethyldithiocarbamate** **Methods.** Neutralise 10 ml of the mineralisate with ammonium hydroxide to pH 3.0 and shake with 5 ml of plumbum chloroform diethyldithiocarbamate solution until the chloroform layer is coloured. Next, re-extract with a 6M hydrochloric acid solution. Confirmatory reactions are performed with the re-extract.			
2.	*Independent work* **Reaction with zinc sulphate and ammonium tetrarodanmercurate** **Methods.** Add 0.2 g of zinc sulphate and a few drops of ammonium tetrarodanomercuroate to 1/3 of the aqueous re-extract.			
3.	**Reaction with pyridinrodanine reagent Methods.** To 1/3 of the aqueous reextract, add 1-2 ml of pyridinrodan reagent dropwise until turbidity or precipitation occurs, then 1-2 ml of chloroform.			
4.	**<u>Reaction of copper and cadmium</u> ferrocyanide formation** **Methods.** Add 10 drops of 2% cadmium chloride solution and 1-2 drops of 5%			

	potassium ferrocyanide solution to 1/3 of the aqueous re-extract.			
1.	**Bismuth detection <u>Reaction of thiourea complex formation</u>** **Methods.** Add 3-5 ml of saturated thiourea solution to a test tube with 5 ml of mineralisate.			
2.	**<u>Reaction of iodobismuthate complex formation with 8-oxyquinoline</u>** **Methods.** Add 20-30 drops of 20% sodium thiosulfate solution to a test tube with 10 ml of mineralisate, then 10 drops of potassium sodium tartrate and excess crystalline potassium iodide until yellow or orange colouration is formed, then carefully add 1-2 ml of 2% 8-oxyquinoline solution in 5% hydrochloric acid solution along the walls.			
1.	**Zinc detection Preliminary reaction with dithizone Methods.** Add 0.25 ml of saturated sodium thiosulfate solution to 0.5 ml of mineralisate, adjust to pH 5-5.5, add 1 ml of acetate buffer, 2 drops of 0.01% dithizone solution in chloroform and 1 ml of chloroform.			
2.	**<u>Separation of zinc from mineralisation and basic reactions</u>** **Methods.** Add 4 ml of potassium sodium tartrate or 20% citric acid solution, 1 ml of saturated thiourea solution or sodium thiosulfate to 10 ml of mineralisate, add a few drops of Nile blue and 2,5M potassium hydroxide solution until pink colouration appears, followed by 1M sulfuric acid solution to pH 8.5 according to the universal indicator paper, then add 3 ml of 1% potassium diethyldithiocarbamate solution, 5 ml of chloroform and shake the mixture vigorously. The chloroform extract is separated, washed with water, and shaken with 3 mL of 1M hydrochloric acid for 30 s. The aqueous layer (reextract) is tested for zinc.			

3.	**Reaction for the formation of zinc hexacyanoferrate (II)** **Methods.** To 1 ml of the test solution add a solution of potassium hydroxide to pH 5.0 (according to the universal indicator paper) and 3-4 drops of 5% potassium hexacyanoferrate (II) solution.			
4.	**Zinc sulphide formation reaction** **Methods.** Add potassium hydroxide solution to pH 5.0 and 2-3 drops of freshly prepared sodium sulfide solution to 1 ml of the test solution.			
5.	*Independent work* **Reaction for the formation of zinc tetrarodanomeric acid** **Methods.** Apply 3-4 drops of the re-extract to a slide and evaporate, dissolve the remainder in a 10% acetic acid solution and add a drop of ammonium tetrarodanomercuroate.			
1.	**Detection of stibium Preliminary reaction with malachite or diamond green** **Method.** Place 5 ml of the mineralisate in a separating funnel, add 1 ml of concentrated sulfuric acid, 3 ml of 5 M hydrochloric acid solution, 2 drops of 5% sodium nitrite solution, and after 5 min - 1 ml of saturated urea solution, 7 drops of 0.5% malachite green solution, 2 g of anhydrous sodium sulfate and 5 ml of toluene. Shake the mixture for 10-25 seconds.			
2.	**Reaction with sodium thiosulfate** **Methods.** Add 5 drops of saturated sodium thiosulfate solution to 5 ml of mineralisate and boil the solution for 1-2 minutes.			
1.	**Arsenic detection Sanger-Black preliminary test Methods.** Into the flask of the Sanger- Black apparatus are added in sequence: 2 mL of mineralisate, 10 mL of 2M sulfuric acid solution, 5 mL of water, 1 mL of 10% tin (II) chloride solution in			

	sulfuric or hydrochloric acid, and then 2 g of purchased zinc. The flask is closed with a nozzle into which reactive paper impregnated with a solution of mercuric bromide or chloride and dried is placed; a cotton swab treated with plumbum acetate is inserted between the paper and the reaction mixture. Observe a yellow, brown or brownish-brown spot on the paper immediately or after 45 minutes in the presence of arsenic. If the stain does not appear after 45 minutes, the paper is first dipped in a 3% potassium iodide solution until the entire surface is reddened, then in a saturated potassium iodide solution until the red colour disappears - a brown stain appears in the presence of arsenic.			
2.	*Independent work* **Tests in the Marsh apparatus Methods.** The flask of the Marsh apparatus is filled in sequentially: 10 g of purchased zinc, and 10-20 ml of 2M sulfuric acid solution is added through a separating funnel soldered into the flask. Check the tightness of the device, and then the completeness of air displacement using the effect of ignition of the released gas collected in the test tube (when air and hydrogen are mixed, crackling, when air is displaced by hydrogen, no sound). When the air is completely displaced from the Marsh apparatus by hydrogen, a narrow part of the Marsh tube is heated using a gas burner flame, and the tube section is cooled at the heating point. Through a funnel, 20 mL of the mineralisation mixed with 2 mL of a 10% solution of tin (II) chloride is added dropwise to the flask of the Marsh apparatus for 30-40 min. Observe in the presence of arsenic immediately or after 60 min in the place of cooling of the Marsh tube an arsenic deposit in the form of a greyish-black mirror or shiny brown colour. The device is cooled, the Marsh tube is removed, and the place of the deposit is heated with air			

	at a 45° angle. In the presence of arsenic, characteristic arsenic anhydride crystals (octahedra and tetrahedra) are formed.			

DESTRUCTION OF BIOLOGICAL MATERIAL AND DETECTION OF MERCURY IN THE DESTRUCT.

Destruction of biological material with a mixture of sulphuric and nitric acids Methodology. Place 20 g of the crushed organ sample (liver and kidneys separately) in a conical flask and add 5 ml of water, 1 ml of ethanol and 10 ml of concentrated nitric acid. Then 20 mL of concentrated sulfuric acid is added dropwise, being careful not to add excess to ensure that nitrogen oxides are released from the flask as little as possible. The flasks with the mixture are left for 5 min to stop the release of nitrogen oxides, and then heated in a boiling water bath for 10-15 min. The violent reaction in the flask is temporarily stopped by adding 30-50 ml of hot water, then immediately the destructant is mixed with twice the volume of boiling water and filtered hot through a filter moistened with water into a flask containing 20 ml of saturated urea solution. The filter is rinsed with hot water, connecting the rinsing water to the destructant. The liquid is cooled, diluted to a certain volume with water, and mercury is determined in half the volume of the destructate.

No. p/n	Methods of chemical reactions	Observations.	Chemistry of reactions	Chemical and toxicological assessment of reactions
1.	**Mercury detection Reaction with dithizone** **Methods.** Shake half of the resulting destructate in a separating funnel with 10 ml of chloroform. Discard the yellow chloroform layer. The operation is repeated until a colourless chloroform extract is obtained. Add 10 ml of a 10% solution of hydroxylamine sulphate or ascorbic acid, 5 ml of chloroform and 0.5 ml or 0.01% solution of freshly purified dithizone to the aqueous layer. Shake the mixture vigorously for 30 seconds.			
2.	**Reaction with suspended copper (I) iodide (verification study)** **Methods.** To 3 ml of the destructant add 3 ml of copper iodide (I) suspension.			

_____SESSION NO.

Date.

SUBJECT: *QUANTIFICATION OF MERCURY IN DESTRUCTATE, ARSENIC, ZINC, COPPER, MANGANESE, CHROMIUM, PL UMB UM IN MINERALISA TE*

QUANTIFICATION OF PLUMBUM PHOTOELECTROCOLOURIMETRIC METHOD

Methodology. Measure the optical density of plumbum dithizonate, obtained as described in the section "Detection of plumbum", on a photoelectrocolourimeter at 520 nm in a cuvette with an absorbing layer thickness of 10 mm. The reference solution is chloroform. The amount of plumbum is then determined from the calibration graph.

Drawing a calibration graph. Prepare a standard solution from recrystallised plumbum nitrate salt with a metal content of 1 mg/ml. Use plumbum solutions of 0.001 and 0.1 mg/ml to construct the calibration graph. Measure out the volumes of solution corresponding to 0.001, 0.002, 0.01, 0.02, 0.05 mg plumbum and perform the extraction as described in the section "Detection of plumbum". The extracts are diluted to 10 ml with chloroform and the optical density is measured.

Formula for calculation:

$$x = \frac{c*Ve*100}{n}$$

x - amount of plumbum per 100 g of organ, mg;

c is the concentration of plumbum in the extract, mg/ml;

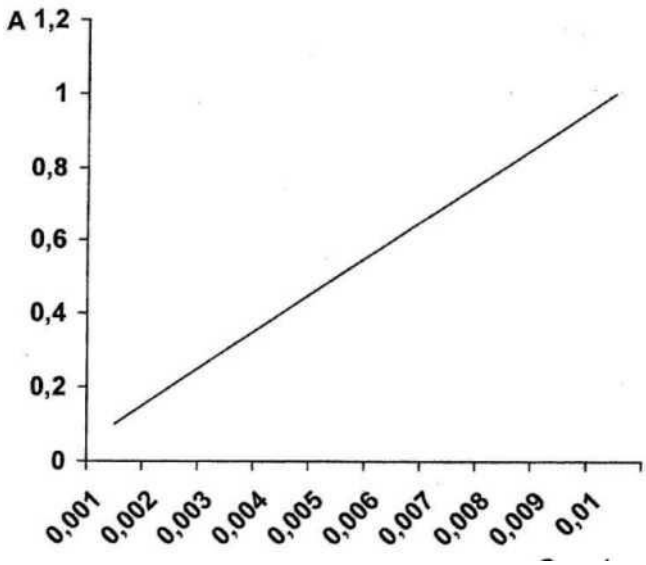
Ve is the volume of dithizone plumbum extract, ml;

n - organ weight, g.

Sensitivity of the quantification method.

Plumbum is determined by photoelectrocolourimetry in the range of 0.02-2 mg per 100g of organ.

Graduation chart for the quantitative determination of plumbum



INDEPENDENT WORK

QUANTIFICATION OF ARSENIC
COLOURIMETRIC METHOD (SANGER-BLACK)

Methodology. A Sanger-Black flask is filled with 0.5-2.0 ml of mineralisate or a solution of mineralisate diluted 5-10 times (for the study of large amounts of arsenic - 0.5-1 mg per 100 g of organ), 10 ml of 2M sulfuric acid solution, 5 ml of water, 1 ml of 10% tin (II) chloride solution, 2 g of activated zinc. The flask is covered with a nozzle, into which a cotton swab treated with plumbum acetate is inserted, and a paper impregnated with mercuric bromide is placed on a horizontal surface with a small hole in the nozzle. After 45 minutes, the reactive paper is removed from the nozzle with tweezers and the resulting stain is compared with a standard scale. The determination is carried out in three different volumes of mineralisation, and the average result is used to draw a conclusion about the amount of arsenic.

Preparation of the standard scale. Prepare a standard solution of arsenic containing 1 mg/ml in 2M sulfuric acid solution. To do this, purify the arsenic trioxide by sublimation. Dissolve the standard in a small amount of 2M potassium hydroxide solution and add 2M sulfuric acid solution to the required volume. A standard scale is prepared from the standard solution before use in the range of $1*10^{-4}$ - $1*10^{-3}$ mg of arsenic.

Formula for calculation:

$x = \frac{a*V*100}{Vi*n}$, where:

x - amount of arsenic per 100 g of organ, mg;
a is the amount of arsenic in the sample to be analysed, mg;
V is the amount of mineralisation after destruction, ml;
Vi is the volume of mineralisation taken for determination, ml; n - organ weight, g.

Sensitivity of the quantification method.

Arsenic is determined by the Sanger-Black method in the range of 0.01-1 mg per 100 g of organ.

____SESSION NO.

Date.

TOPIC: *A GROUP OF SUBSTANCES ISOLATED FROM BIOLOGICAL MATERIAL BY STEAM DISTILLATION - VOLATILE POISONS. METHODS OF ISOLATING VOLATILE POISONS FROM BIOLOGICAL MATERIAL.*

DETECTION OF VOLATILE POISONS BY CHEMICAL METHODS (HYDROCYANIC ACID, FORMALDEHYDE, HYDROCARBON HALIDES).

Method of isolating volatile poisons. Assemble a water vapour distillation apparatus. Pour water into the steamer and bring it to a boil. While the water in the steamer is heating, take 20-100 g of

the crushed herbage, place it in a distillation flask, mix it with water to a thick slurry, but so that the mixture occupies no more than 1/3 of the flask volume (to prevent overflow). The distillation flask with the object is placed in a cold water bath, acidified with a 10% solution of tartaric or oxalic acid to pH 2-3, quickly closed with a stopper, connected to a refrigerator and carefully (sharp steam) to a steam generator. Continue heating the steamer and simultaneously turn on the water bath heating.

The first portion of the distillate (3 mL) is collected in a receiver with 2 mL of 5% sodium hydroxide solution to capture hydrocyanic acid. Empty flasks with 25 mL marks are prepared for the discharge of subsequent portions. The distillation is carried out at such a speed that the drops in the receiver can be counted. For very fast distillation, reduce the flame of the burner under the steam generator, and vice versa for slow distillation.

For the ethylene glycol test, the distillate is collected in a volume of 500 ml (young compounds).

Isolation with acetic acid is carried out by acidifying the objects with a 10% solution of sulfuric or orthophosphoric acid, and the distillate is collected in a receiver containing a 0.1M sodium hydroxide solution.

When analysing objects for methanol, the distillate is collected in a cooled receiver.

For the quantitative determination of volatile poisons, distillation leads to a negative reaction to the substances to be analysed.

DETECTION OF VOLATILE POISONS BY CHEMICAL METHODS (HYDROCYANIC ACID, FORMALDEHYDE, HYDROCARBON HALIDES).

No. p/n	Methods of chemical reactions	Observations.	Chemistry of reactions	Chemical and toxicological assessment of reactions
1.	*STUDY OF DISTILLATE NO. 1* **Detection of hydrocyanic acid** **Reaction of the formation of Berlin <u>blue</u>** **Methods**. To 1 ml of alkaline distillate add 2-3 drops of a 40% solution of ferric (II) sulphate containing traces of ferric			

	(III). The mixture is shaken, heated to almost boiling, and then cooled to room temperature and a 10% hydrochloric acid solution is added until the reaction is slightly acidic.			
1.	*STUDY OF DISTILLATE NO. 2* **Formaldehyde detection <u>Reaction with alkaline resorcinol solution</u>** **Methods**. Add 1 ml of freshly prepared reagent to 1 ml of the test solution: 1% resorcinol solution is mixed with an equal volume of 10% sodium hydroxide solution. A "blind" experiment with the reagents is performed in parallel. The contents of both tubes are heated for 3-5 min in a water bath.			
2.	**Reduction of copper hydroxide with <u>Fehling's reagent</u>** **Methodology.** Add 2 drops of sodium hydroxide solution to 1 ml of the test solution until alkaline and 2-3 drops of Fehling's reagent (Fehling's reagent No. 1 + Fehling's reagent No. 2 (1:1)). Stir the contents of the test tube and heat in a water bath.			
3.	**Reaction with codeine sulfate** **Methods.** In a porcelain cup, mix 1 ml of the test solution with 5 ml of concentrated sulfuric acid. After cooling, add a crystal of codeine.			
4.	**Reaction with chromotropic acid** **Methodology.** 1 ml of the test solution is placed in a porcelain cup, 3-4 drops of concentrated chromatrophic acid are added and mixed gently.			
5.	**Reaction with fuchsinic acid** **Methods.** Add 2-3 drops of concentrated sulfuric acid to 1 ml of the test solution. The contents of the test tube are stirred and cooled, and then 1 ml of fuchsin- sulfuric acid is added.			
	Detection of halogenated hydrocarbons (chloroform, chloral			

1.	**hydrate, carbon tetrachloride, 1,2-dichloroethane)** **<u>Reaction for the cleavage of</u> organically bound chlorine** **Methods.** To 1-2 ml of the test solution is added 1 ml of a 10% alcohol solution of sodium hydroxide. The test tube with the contents is heated in a boiling water bath for 30 min or over bare fire for 3-5 min. Cool the solution, add 10% nitric acid solution to an acidic reaction, 0.5 ml of 1% argentium nitrate solution.			
2.	*Independent work* **Formation of isonitrile** **Methods.** To 1 ml of the test solution add 10 drops of a 10% alcohol solution of sodium hydroxide and a drop of an aqueous solution of aniline. The liquid is heated in a water bath for 1-2 min. To decompose isonitrile, add a 10% solution of sulfuric acid to the test tube and boil the solution until the odour disappears.			
3.	**Reaction with alkaline resorcinol <u>solution</u>** **Methods.** Add 1 ml of freshly prepared reagent to 1 ml of the test solution: 1% resorcinol solution is mixed with an equal volume of 10% sodium hydroxide solution. A "blind" experiment with the reagents is performed in parallel. The contents of both tubes are heated for 3-5 min in a water bath.			
4.	**Reaction with Fehling's reagent** **Methods.** To 1 ml of the test solution add 2 drops of 10% sodium hydroxide solution until alkaline and 2-3 drops of Fehling's reagent (Fehling's reagent No. 1 + Fehling's reagent No. 2 (1:1)). Stir the contents of the test tube and heat in a water bath.			
5.	**Reaction with Nessler's reagent** **Methods.** Add 2-3 drops of Nessler's reagent to 0.5 ml of the test solution and mix.			
	Independent work			

6.	**Detection of 1,2-dichloroethane <u>Reaction with fuchsin sulfuric acid (after conversion of 1,2-dichloroethane to formaldehyde)</u>** **Methods.** Pour 0.5 ml of the test solution and 0.5 ml of 10% sodium carbonate solution into a 1 ml ampoule. The ampoule is sealed and heated in a boiling water bath for 1-2 hours. After cooling, the contents of the ampoule are transferred to a test tube, where a 10% solution of sulfuric acid is added until the acid reaction is observed by the universal indicator paper, and then 2 drops of a 5% solution of potassium periodate in 2M sulfuric acid solution. After 5 min, the presence of formaldehyde in the solution is determined by reaction with fuchsin- sulfuric acid. Add 2-3 drops of concentrated sulfuric or hydrochloric acid to 1 ml of the test solution. The contents of the test tube are stirred and cooled, and then 1 ml of fuchsinic acid is added.			
7.	**<u>Reaction for the formation of copper acetylenide</u>** **Methods.** Into a 1 ml ampoule add 0.5 ml of distillate and 0.5 ml of 30% sodium caustic solution. The ampoule is sealed and heated for one hour. After that, the ampoule is cooled, opened, and its contents are transferred to a test tube to which 30% acetic acid solution is added until the litmus test is acidic. To this liquid is added 2 drops of freshly prepared ammonia solution of copper (I) salt.			

Results of qualitative reactions for the detection of halogenated hydrocarbons

TABLE 1

No. p/n	**Reactions.**	**Test substances**

		chloroform	chloral hydrate	carbon tetrachlorid e	1,2-dichloroet hane
1.	**Chlorine separation**				
2.	**Formation of isonitrile**				
3.	**With alkaline resorcinol solution**				
4.	**With Fehling's reagent**				
5.	**With Nessler's reagent**				

____SESSION NO.

Date.

TOPIC: *DETECTION OF VOLATILE POISONS BY CHEMICAL METHODS (SINGLE-ATOMALCOHOLS, ACETONE, PHENOL, ACETIC ACID).*

No. p/n	Methods of chemical reactions	Observations.	Chemistry of reactions	Chemical and toxicological assessment of reactions
1.	***Detection of single-atom alcohols*** **Methanol detection** **Reaction for the formation of salicylic acid methyl ester.** **Methods.** Pour 1 ml of the distillate into a test tube, add 0.03-0.05 g of salicylic acid, and then heat the mixture gently.			

2.	**Oxidation of methanol to formaldehyde.** **Methods.** Add 2-3 ml of 10% sulfuric acid solution to 5 ml of the test solution, cool the liquid and add 1% potassium permanganate solution to it until it turns a stable pink colour, avoiding excess reagent. After 15-20 minutes, the excess oxidant is destroyed with a 15% oxalic acid solution or a 15% sodium sulfite solution. The resulting colourless solution is used to perform formaldehyde reactions (see the topic of formaldehyde detection).			
1.	**Ethanol detection** **Iodoform formation reaction** **Methods.** To 1 ml of the test solution add 2 ml of 5% sodium hydroxide or potassium carbonate solution and dropwise 1% iodine solution in 2% potassium iodide solution until the mixture turns yellow. The mixture is heated in a water bath at 40-50°C.			
2.	**Detection of acetic-ethyl ether** **Methodology.** To 1 ml of the test solution add 0.1 g of dried sodium acetate, then double the volume of concentrated sulfuric acid. The mixture is heated in a boiling water bath until gas bubbles are released.			
3.	*Independent work* **Reaction for the formation of ethyl benzoate** **Methodology.** Mix 1 ml of the test solution with 1-2 drops of benzoyl chloride and add 10% caustic sodium solution dropwise with frequent shaking.			

1.	**Detection of isopentanol <u>The reaction of isoamyl acetate formation.</u>** **Methods.** The ether extract from part of the distillate is evaporated to remove the ether, mixed with two drops of concentrated sulfuric acid and a small amount (0.003 g) of dried sodium acetate. The mixture is heated in a water bath.			
2.	*Independent work* **Reaction with salicylic aldehyde** **Methods.** The ether extract from a part of the distillate is evaporated to remove the ether. To the residue add 1 ml of 1% salicylic aldehyde solution and 3 ml of concentrated sulfuric acid. After cooling, the mixture in the test tube is heated in a boiling water bath for 3 min.			
3.	**Reaction with a solution of paradimethylaminobenzaldehyde in concentrated sulfuric acid** **Methodology.** Add 5-10 drops of reagent (para-dimethylaminobenzaldehyde in concentrated sulfuric acid) to the residue.			
1.	**Acetone detection Iodoform formation reaction** **Methods.** Add 1 ml of 10% ammonium hydroxide and a few drops of iodine solution in potassium iodide to 1 ml of the test solution.			
2.	**Reaction with nitroprusside** **Methods.** Add 1 ml of 10% sodium hydroxide solution and a few drops of 1% freshly prepared sodium nitroprusside solution to 1 ml of the test solution.			
	Phenol detection **Preparation of the distillate for analysis** **Methods. A** part of the third distillate is alkalised with sodium bicarbonate to an alkaline reaction, transferred to a separating funnel and the phenol is extracted with diethyl ether (2-3 times 10			

1.	ml each). Combine the ether extracts and evaporate to dryness at room temperature. The residue is dissolved in 1-2 ml of water and examined. **Reaction of tribromophenol formation** **Methods.** Add 2-4 drops of saturated bromine water solution to 0.5-1 ml of the test solution.			
2.	**<u>Reaction with ferric (III) chloride</u>** **Methods.** Place 1 drop of the test solution in a porcelain cup, then 1 drop of a freshly prepared solution of ferric chloride (III). A "blind" experiment is carried out nearby.			
1.	**Detection of acetic acid** **Reaction with ferric (III) chloride** **Methods.** To 2-3 ml of the test solution add 1 drop of a freshly prepared 5% solution of ferric (III) chloride. A "blind" experiment is carried out nearby.			
2.	**<u>Reaction for the formation of acetyl ethyl ester</u>** **Methods.** Mix 1 ml of the test solution with 1 ml of ethanol and 2 ml of concentrated sulfuric acid. The test tube with the solution mixture is heated in a boiling water bath.			

_____SESSION NO.

Date.

TOPIC: *ANALYSIS OF DISTILLATE AND BIOLOGICAL FLUIDS (BLOOD, URINE) FOR VOLATILE POISONS BY GAS-LIQUID CHROMATOGRAPHY (GLC).*

DETECTION AND QUANTIFICATION OF ETHANOL IN BIOLOGICAL FLUIDS BY GC METHOD

<u>Check the speaker resolution and detector sensitivity.</u>

Methods. Pour 0.5 ml of 50% trichloroacetic acid and 0.5 ml of the test mixture into a penicillin vial. The vial is closed with a rubber stopper that is fixed. The contents of the vial are stirred and 0.5 ml of 30% sodium nitrate solution is injected through the stopper using a syringe. The contents are stirred again (30 pendulous or circular movements on the table surface) and left for 1 min. After that, 0.5-3.0 ml (depending on the sensitivity of the detector, but always the same amount) of the vapour sample is taken from the vial using a syringe and injected into the chromatograph. The absolute and relative (relative to propanol) retention times of the test mixture ingredients are determined. When

measuring the relative retention time, it is recommended to count from the maximum peak of air to the maximum peak of the substance to be determined (corrected retention time - t'R) rather than from the point of sample injection into the chromatograph.

DETECTION OF ALCOHOLS IN ANALYSED SAMPLES (BLOOD, URINE, DISTILLATE)

Methodology. Pour 0.5 ml of 50% trichloroacetic acid and 0.5 ml of the liquid to be analysed into a penicillin vial. The vial is closed with a stopper, which is secured with a special fixative. Proceed as described for checking the column resolution and detector sensitivity. Compare the retention time of the test and the sample to be analysed. If ethanol is detected, it is quantified.

Quantification of ethanol

Construction of a calibration graph. Prepare a series of standard ethanol solutions of 2.3, 4.5% and internal standard solutions containing 4% propyl alcohol. In several penicillin vials, add 2 ml of internal standard and 2 ml of ethanol solution of different concentrations (2.3.4.5%). Mix the contents of the vials well, and then take 1 ml from each vial and transfer to other penicillin vials. Add 0.5 mL of 50% trichloroacetic acid solution to each vial and proceed as described above. Measure the heights of the ethinitrite and propyl nitrite peaks. Find the ratio of these values, which is multiplied by 100, and then by 0.95 (correction factor for blood) and 1.05 (correction factor for urine). Based on the data obtained, graphs of the ratio of peak heights versus the concentration (in ppm) of ethanol in

No. p/n	%		h of ethanol (mm)	h of propanol (mm)	h ethanol/ h propanol	coefficients (R)	
	ethanol	propanol				0,95 (blood)	1,05 (urine)
1.	1	4	14,0	49,0			
2.	2	4	35,5	50,5			
3.	3	4	56,0	52,0			
4.	4	4	82,5	50,5			

standard solutions of blood and urine are plotted.TABLE 1

Graduated schedule according to Table 1

Graduated schedule according to Table 1

_____SESSION NO.

Date.

TOPIC: *GROUP OF SUBSTANCES ISOLATED BY ORGANIC SOLVENTS (PESTICIDES). METHODS OF ISOLATION OF PESTICIDES FROM BIOLOGICAL OBJECTS AND STUDY OF EXTRACTS.*

No. p/n	Methods of chemical reactions	Observations.	Chemistry of reactions	Chemical and toxicological assessment of reactions
1.	**Biochemical sample** **Methodology.** Take two test tubes, add 1 ml of the test solution to one tube and 1 ml of pure water (twice distilled) to the			

	other tube. Add cholinesterase solution dropwise to both test tubes and mix. Then add acetylcholine, mix again and then one drop of bromothymol blue indicator-buffer solution. Compare in test tubes.			
1.	**Chemical analysis of chlorophos** **General reactions Detection of phosphate ions Methods.** Pour 3-5 drops of the mineralisate into a test tube and add 5 drops of ammonium molybdate solution. The mixture is acidified with a 10% solution of nitric acid. To this solution add 3-5 drops of a saturated solution of benzidine hydrochloride. Then add 10% ammonia solution until the reaction is alkaline (litmus test).			
2.	*Independent work* **Hydroperoxide test** **Methods.** Add 2 ml of the test liquid to a mixture of 0.5 ml of 2.5% acetated benzidine solution and 2 ml of freshly prepared hydrogen peroxide in alkaline medium.			
3.	*Frequent reactions to chlorophos* **Reaction with Nessler's reagent** **Methodology.** Add 2-3 drops of Nessler's reagent to 0.5 ml of the test solution and mix the liquids.			
4.	**Reaction with alkaline resorcinol solution** **Methodology.** Add an equal amount of a mixture consisting of 1% resorcinol solution and 10% sodium hydroxide solution, prepared before use, to 0.5-1 ml of the test water.			
5.	**Analysis of chlorophos by thin sorbent layer chromatography (TLC)** A drop of the extract to be tested and a drop of the "witness" solution are applied to the Silufol plate. The plate is placed in a chromatographic chamber with a hexane-acetone (1:1) solvent system. The plate is dried and developed with a			

	freshly prepared mixture of solutions of 2% resorcinol and 10% sodium carbonate (2:3).			

AN APPROXIMATE ACT OF FORENSIC CHEMICAL (CHEMICAL AND TOXICOLOGICAL) EXAMINATION

ACT NO.
FORENSIC CHEMICAL EXAMINATION
INTERNAL ORGANS OF THE CORPSE OF G.H., SENT TO THE INVESTIGATOR

The order for the appointment of a forensic chemical examination from
, the act of forensic medical examination of the corpse from and other correspondence on the letters.

The study was conducted at by an expert
chemist at.
Started Completed

CIRCUMSTANCES OF THE CASE

Gr.H. in a state of intoxication ""On 20 _____ , between 19 and 21 hours, he consumed powders of unknown composition, after which he soon lost consciousness. He was taken by ambulance to the hospital at 23 hours, where he died at 23.30 hours without regaining consciousness.

EXTERNAL INSPECTION

The following items were delivered for examination: a 750 ml bio-glass jar No. 1, covered with parchment folded in half. The mouth of the jar was closed with a cork stopper, wrapped in white paper and tied with a piece of bandage, the ends of which were imprinted on cardboard with a light brown plasticine seal with an unclear imprint of "courts". There is a paper label on the jar with an inscription in purple ink and a seal, the imprint of which is unclear. The inscription on the label reads "Jar No. 1 - stomach contents of the corpse of Mr Kh." The weight of the contents of the jar is 500 g. The contents of the jar are a mushy pinkish-grey mass. The reaction of the medium to the alacrine is acidic. Congo paper is not coloured.

Jar No. 2. A 1000 ml white glass jar covered with parchment and white paper. The mouth of the jar is wrapped with a piece of bandage, the ends of which are imprinted on the cardboard with a light brown plasticine seal with the indistinct inscription "should be used". The jar has a label on white paper with the words in purple ink "Jar No. 2 - pieces of liver and kidneys". The contents of the jar are pieces of the specified organs weighing: liver - 500g, kidneys - 300g. The reaction of the contents of

the jar to the litmus test is neutral. The smell is unremarkable.

A 20 ml test tube, closed with a stopper and filled with blood to the stopper. The test tube has a label with the words "Blood from the corpse of Mr X" written in purple ink.

CHEMICAL ANALYSIS

I. 100 g of the contents of the jar marked No. 1 were mixed with distilled water to a mushy state, acidified with oxalic acid to a slightly acidic reaction to the litmus test and subjected to distillation with water vapour. The first portion of the distillate in the amount of 3 ml was collected in a 5 ml 1% sodium hydroxide solution, the next portions of the distillate of 25 ml were collected in two receivers without sodium hydroxide.

Study of distillates: 1) a few drops of ferrous sulphate and 1-2 drops of ferric oxide chloride solution were added to the entire first distillate, then the liquid was acidified with hydrochloric acid to a sweet acid reaction. No blue precipitate or blue colouration was observed after 48 hours; 2) a solution of resorcinol in caustic soda was added to a part of the second distillate. When heated in a water bath, no colouration was observed; 3) iodine solution in caustic soda was added to a part of the second distillate. When heated in a water bath, a yellow precipitate with the smell of iodoform fell out; 4) an equal volume of 20% sulfuric acid solution and a small amount of potassium permanganate (dry) were added to a part of the second distillate. When standing, a pleasant smell resembling the smell of fruit was felt. After 20 minutes, the liquid was filtered off and the following reactions were performed with the colourless filtrate: a) five times the volume of concentrated sulfuric acid, in which several grains of codeine were dissolved, was added to half of the filtrate. No bluepurple colouration was observed; b) an equal volume of fuchsinic acid and 1 ml of concentrated sulfuric acid were added to the other half of the filtrate. No colouration was observed; 5) a few grains of sodium acetate were dissolved in 1 ml of the second distillate, the latter was mixed with a double volume of concentrated sulfuric acid. When heated slightly, the smell of acetic-ethyl ether was felt; 6) a part of the second distillate was mixed with an equal volume of 5% alcohol solution of caustic soda and gently heated for a long time, after cooling, the liquid was acidified with nitric acid and a 5% solution of argentium nitrate was added to it. There was no formation of turbidity or precipitate. The residue of the second distillate was mixed with the third and subjected to examination; 7) a few drops of bromine water were added to some of the distillates, immediately forming a cloudy whitish- yellow colour; half of the mixture of distillates was subjected to two times deflemmation. After the second deflagration, the first distillate was collected with 5 ml of liquid, and the second with 10 ml of liquid. With the first distillate, the reactions described in this section under eqs. 3, 4, 5, with the same results, with the reaction described in step 5, the smell of acetyl ether was clearer. No turbidity was observed during the reaction with bromine water; 8) the second part of the mixture of distillates was combined together, alkalised with sodium carbonate solution and re-extracted with ether. The ether extracts were combined together, filtered, and the ether was removed at room temperature. The residue was insignificant. After dissolving it in 2-3 drops of distilled water, 2 drops of 5% freshly prepared ferric oxide chloride solution were added to the resulting solution. No purple colouration was observed.

II. 100 g of the contents of the jar designated by us as No. 1 were crushed, combined together, poured with ethyl alcohol, acidified with an alcoholic solution of oxalic acid to a slightly acidic reaction by litmus test. The next day, the alcohol was poured off, and the remaining object was again poured with ethyl alcohol, slightly acidified with an alcoholic solution of oxalic acid, and left for a day at room temperature.

This operation was repeated again. Then all alcohol extracts were combined together, evaporated at 40° to a syrupy liquid, which was treated with a small amount of alcohol. The coagulated proteins were filtered off, and the liquid was evaporated again at 40°. This operation was repeated until no flakes were observed when alcohol was added. After that, the syrupy residue was treated with 25 ml of distilled water and the aqueous solution was extracted again with chloroform. The chloroform extractions were combined together, filtered and the chloroform removed at room temperature. The residue after removal of the chloroform from the acid chloroform extraction was brown in colour and oily. After treating this residue with 20 ml of hot distilled water alkalised with caustic soda, the chloroform extraction was repeated. The alkaline chloroform extractions were combined together and the chloroform was removed at room temperature. Bromine water was added to the residue; the liquid was evaporated to dryness in a water bath. After evaporation, the dry residue was wetted with a drop of concentrated ammonia solution, and a purple-red colouration was observed at the edges. The aqueous liquid of the alkaline reaction was acidified with sulfuric acid and re-extracted with ether. The ether extractions were combined together, filtered and the ether was removed at room temperature. The slightly brown, oily residue was dissolved in a small amount of ether and distributed on several slides. The ether was removed at room temperature. The following reactions were performed with the residue: 1) a drop of concentrated sulfuric acid was applied to the residue, followed by a drop of distilled water. A white amorphous precipitate appeared, which turned into a crystalline precipitate in an hour, spherical growths of needle crystals formed at the edges of the drop; 2) a drop of ammonia was applied to the residue, followed by a drop of 10% hydrochloric acid solution. A white amorphous precipitate appeared, crystallising after 30 minutes with the formation of similar growths at the edge of the drop; 3) a drop of ferric ferric reagent was applied to the residue, and after 20 minutes, growths of dark brown needle crystals formed at the edges of the drop; 4) one drop of concentrated sulfuric acid and a small crystal of potassium bichromate were applied to the residue. When the crystal moved, a yellow-green colouration was observed; 5) a drop of concentrated nitric acid was applied to the residue; no colouration was observed; 6) a drop of concentrated sulfuric acid containing formalin was applied to the residue; no colouration appeared either once or when it was standing. After extraction with chloroform, the acidic aqueous solution was alkalised with ammonia and extracted again with chloroform. The alkaline chloroform extractions were combined, filtered, and the chloroform was removed at room temperature. The residue after chloroform removal was colourless and insignificant. After dissolving in a small volume of chloroform, it was distributed on 3 watch glasses. After removing the solvent, the residue was dissolved in 0.1 n hydrochloric acid. The following reactions were performed with the solutions: 1) a slight precipitate appeared
when a drop of bismuth iodide solution was added to potassium iodide; 2) a slight precipitate was observed when a drop of iodine solution was added to potassium iodide; 3) a slight white turbidity appeared when a drop of mercuric iodide solution was added to potassium iodide. A part of the chloroform extraction was distributed into several porcelain cups and after removing the chloroform, the following reactions were carried out with the residue: 4) a drop of concentrated sulfuric acid containing formalin was applied to the residue. No blue-purple colouration was observed either at once or when standing; 5) a drop of concentrated sulfuric acid containing nitrate was applied to the residue. No colouration was observed either at that time or when standing; 6) a drop of concentrated sulfuric acid containing molybdenum acid was applied to the residue. No colouration was observed; 7) a drop of sulphuric acid containing vanadic acid was applied to the residue. Colouration also appeared; 8) a drop of concentrated sulfuric acid was applied to the residue. No purple streams were observed

during the movement of the potassium bichromate crystal; 9) a drop of 10% hydrochloric acid solution was applied to the dry residue on the slide, the drop was dried at room temperature, and then a drop of 1% potassium permanganate solution was applied to the residue; no pinkish square plates were observed; 10) 5 ml of concentrated nitric acid was placed in a cup with the residue, and after evaporation in a water bath, a few drops of an alcohol solution of potassium permanganate and 1 ml of acetone were applied to the dry residue. No purple colouration was observed.

III. 100 g of the contents of the jar marked No. 1 were crushed, poured into 50 ml of nitric acid, diluted twice, and 25 ml of concentrated sulfuric acid. After a vigorous reaction, the mixture was heated with the constant addition of nitric acid dropwise to obtain a colourless liquid that did not darken when heated for 30 minutes without the addition of nitric acid while sulphur oxides were released. An equal volume of distilled water was added to the cooled liquid and formalin was added dropwise until a drop of the test liquid no longer gave a blue colour with a solution of diphenylamine in concentrated sulfuric acid. The mineralisate was diluted with water to 200 ml and examined by the following reactions: 1) 4 ml of water, 1 ml of saturated solution of monosubstituted sodium phosphate, 0.2 g of potassium periodate were added to 1 ml of mineralisate and the liquid was heated for 20 minutes. No pink colouration was observed; 2) 4 ml of water, 1 drop of 10% argentium nitrate solution, 0.5 g of ammonium persulfate were added to 1 ml of mineralisate, and the reaction mixture was heated for 20 minutes in a water bath, then 1 ml of saturated sodium monosubstituted phosphate solution was added to the colourless liquid, pH was adjusted to 1.7 by adding 10% potassium permanganate solution, and 1 ml of diphenylcarbazide solution was added. No colouration was observed; 3) 5 ml of chloroform was added to 5 ml of mineralisate, a few drops of 0.01% dithizone and chloroform solution, and the liquid was shaken vigorously. No golden yellow colouration appeared either immediately or after washing the chloroform layer with a 0.1% solution of ammonium oxide hydrate; 4) 10 ml of the mineral was neutralised with ammonia to pH 3 (according to the universal indicator) and shaken with 5 ml of a chloroform solution of plumbum diethyldithiocarbamate. The chloroform layer was neither yellow nor brown; 6) 1 mL of the mineralisate was placed in a separating funnel, 4 mL of 40% sulfuric acid solution, 3 mL of 5 and hydrochloric acid, 2 drops of 5% sodium nitrate solution, 7 drops of 0.5% alcohol solution of malachite green, 1 g of anhydrous sodium sulfate, 5 mL of toluene were added to it and the liquid was shaken vigorously; neither the aqueous layer nor the toluene layer was coloured; 7) 2 ml of the mineralisate was placed in a Sanger-Black flask, and 10 ml of 4 n sulfuric acid solution, 5 ml of water, 1 ml of 10% tin chloride (P) solution in concentrated sulfuric acid and a controlled 2.0 g of cupric zinc were added. The flask was closed with a stopper with a built-in nozzle, between the bars of which there was a mercuric bromide-treated and dried reactive paper. Below the reactive paper in the neck of the nozzle was a cotton swab treated with plumbum acetate solution and dried. After one hour, the reagent paper did not change colour; 8) 20 drops of 20% sodium thiosulfate solution were added to 10 ml of mineralisate until the purple colouration formed and disappeared, 10 drops of potassium sodium tartrate and an excess of crystalline potassium iodide until orange colouration formed. Then a few drops of a 2% solution of oxyquinoline in 5% hydrochloric acid solution were added. No orange-red precipitate or colouration was observed. When 1 mL of a mixture of acetone and amyl acetate (1:1) was added to the reaction liquid and shaken vigorously, the organic solvent layer did not stain; 9) 10 mL of mineralisate was placed in a separating funnel and 2 mL of glycerol solution (1:10), 4 mL of 10% sodium potassium tartrate solution, 2 drops of Nile blue solution and 10% caustic potassium solution were added to it until a pink colour appeared. To the coloured liquid was added 2 ml of 1%

sodium diethyldithiocarbamate solution and 10 ml of chloroform, after which the contents of the separating funnel were vigorously stirred for 30 seconds.

The chloroform layer was separated, washed with water and shaken vigorously with 3 mL of 1 n hydrochloric acid for 30 seconds.

The hydrochloric acid solution was separated and tested by the following reactions: a) to 1 ml of the solution was added dropwise 10% caustic soda solution to pH 5 and 3 drops of freshly prepared sodium sulfide solution. Neither turbidity nor yellow precipitate was formed; b) 3 drops of potassium ferrocyanide were added to 1 ml of the test solution adjusted to pH 5 as described in (a). Neither precipitate nor white turbidity appeared; 10) 2 drops of saturated sodium thiosulfate solution were added to 0.5 ml of the mineralisate. Adding 10% caustic soda solution dropwise, the pH was adjusted to 5.0 and 1 ml of acetate buffer (pH 5), 2 drops of 0.01% dithizone solution in chloroform, 1 ml of chloroform were added to the reaction mixture. The liquid was shaken vigorously. The chloroform layer did not turn pink or red- violet.

IV. 100 g each of liver and kidneys in the jar designated as No. 2 were subjected to the tests described in Sections I, II, III with the same results. I, II, III with the same results.

V. 20 g of crushed organs (liver and kidneys separately), which were in the jar marked by us as No. 2, were placed in conical flasks and each object was filled with 10 ml of water, 1 ml of ethanol, 10 ml of concentrated nitric acid and 10 ml of concentrated sulfuric acid. After the release of nitrogen oxides stopped, the flasks were heated in a boiling water bath for 20 minutes. The hot destructates were mixed with double volumes of water and the liquid was separated by filtration. The residue on the filter was washed 3 times with hot water and the wash liquids were added to the main extraction. The cooled destructants were diluted with distilled water in volumetric flasks to a volume of 200 ml. To half of each destructate was added 5 ml of 2.5 n sodium sulfite solution, to 250 ml of copper iodide suspension. The liquids were mixed. The suspension in both flasks remained white. After adding the second half of the destructants to the test solutions, the colour of the suspension did not change.

VI. Into a penicillin vial was placed 0.5 ml of trichloroacetic acid solution, a drop of methyl alcohol solution and 0.5 ml of blood in a test tube. After closing the vial with a stopper, the contents were shaken thoroughly, 0.25 ml of sodium nitrite solution was injected into the vial with a syringe, and the mixture was shaken vigorously again.

Then, after 1 minute of the vial, 3 mL of the vapour phase was withdrawn with a syringe and injected into the gas chromatograph. The ethyl nitrite peak was recorded on the chromatograph.

2 mL of isopropyl alcohol solution (internal standard) was mixed with 2 mL of the same blood, and 1 mL of the mixture was injected into a penicillin vial containing 0.5 mL of trichloroacetic acid solution. After closing the vial with a stopper, the contents were processed as described above.

After injecting 3 ml of the vapour phase into the chromatograph, the chromatogram showed a peak height of ethyl nitrite equal to 72 mm and a height of

The isopropyl nitrite peak was 69 mm, while the repeated determination was 83 and 78 mm, respectively.

Chromatography conditions: chromatograph ХЛ4. column 200x0.6 cm. Inzen brick (0.2-0.3 mm) + 0.3% caustic ether + 25% triethylene glycol (by weight of brick).

The temperature is 70°. Nitrogen carrier gas consumption is 2 l/hour.

From the above, it follows that the chemical and toxicological examination of the internal organs of

the corpse of Gr. H.'s internal organs, the

CONCLUSION

Based on the above, it follows that during the chemical and toxicological examination of the internal organs of the corpse of Mr H., sent for examination, with the decision of
from 200 g Found: ethyl alcohol in the amount of 2.00%,
caffeine and phenobarbital. Not found: methyl alcohol, volatile organic halogenated substances, phenols, formaldehyde and hydrocyanic acid, morphine, codeine, atropine, strychnine, brucine, cocaine, promethol, barium, plumbum, manganese, chromium, argentum, argentum bismuth, cadmium, zinc and mercury.

Supplement: 1) 2 chromatograms on 2 sheets;

2) calibration chart on 1 sheet;
3) 2 photos of microcrystals.

"___"_____ 20__p
Аналіз виробляв(а) _____