TOXICOLOGICAL SIGNIFICANCE, INFLUENCE ON THE ORGANISM, AND METHODS OF DETERMINING CARBON (II) OXIDE

Fozilov Hasan Sadriddin ugli,
Acting Associate Professor of Bukhara State Technical University, Doctor of Philosophy in Technical Sciences
E-mail: fozilovhasann@gmail.com
Tel: (+99891) 440-52-13
Fozilov Sadriddin Fayzullaevich,
Professor at Bukhara State Technical University, Doctor of Technical Sciences
E-mail: sadriddinf@mail.ru
Tel: (+99890) 718-40-66
Rabbimov Jaxongir Shodmonkulovich,
Acting Associate Professor of Karshi State Technical University, Doctor of Philosophy in Technical Sciences
E-mail: rabbimov.jahongir1933@gmail.com
Tel: (+99899) 064-06-38

Abstract. This article discusses the physicochemical properties of carbon monoxide (CO), its toxic effects on the human body, clinical and pathological signs of CO poisoning, and the methods used to detect carboxyhemoglobin in forensic medical practice. The mechanisms of CO binding to hemoglobin, formation of stable carboxyhemoglobin, and development of severe tissue hypoxia are described in detail. Special attention is given to spectroscopic, spectrophotometric, and chemical techniques for the detection of CO intoxication, as well as the common sources and prevalence of carbon monoxide exposure in domestic and industrial environments.

Keywords: Carbon monoxide, CO, carboxyhemoglobin, toxicology, spectrophotometry, spectroscopy, hypoxia, hemoglobin.

Аннотация. Аннотация. В данной статье рассматриваются физико-химические свойства монооксида углерода (CO), его токсическое воздействие на организм человека, клинические и патологические признаки отравления CO, а также методы, используемые для определения карбоксигемоглобина в судебно-медицинской практике. Подробно описаны механизмы связывания CO с гемоглобином, образования стабильного карбоксигемоглобина и развития тяжелой тканевой гипоксии. Особое внимание уделяется спектроскопическим, спектрофотометрическим и химическим методам выявления отравления угарным газом, а также общим источникам и распространенности воздействия угарного газа в бытовых и промышленных условиях.

ISSN 3030-3907

Ключевые слова: монооксид углерода, CO, карбоксигемоглобин, токсикология, спектрофотометрия, спектроскопия, гипоксия, гемоглобин.
Ключевые слова: монооксид углерода, CO, карбоксигемоглобин, токсикология, спектрофотометрия, спектроскопия, гипоксия, гемоглобин.
Annotatsiya. Ushbu maqolada uglerod oksidi (CO) ning fizik-kimyoviy xossalari, uning inson organizmiga toksik ta'siri, CO bilan zaharlanishning klinik va patologik belgilari, sud-tibbiy amaliyotda karboksigemoglobinni aniqlashda qo'llaniladigan usullar haqida so'z boradizoh. Ushbu maqolada uglerod oksidi (CO) ning fizik-kimyoviy xossalari, uning inson organizmiga toksik ta'siri, CO bilan zaharlanishning klinik va patologik belgilari, sud-tibbiy amaliyotda karboksigemoglobinni aniqlashda qo'llaniladigan usullar haqida so'z boradi. CO ning gemoglobin bilan bog'lanishi, barqaror karboksihemoglobin hosil bo'lishi va to'qimalarning og'ir gipoksiyasi rivojlanishi mexanizmlari batafsil tavsiflangan. Co intoksikatsiyasini aniqlash uchun spektroskopik, spektrofotometrik va kimyoviy usullarga, shuningdek, maishiy va sanoat muhitida uglerod oksidi ta'sirining umumiy manbalari va tarqalishiga alohida e'tibor beriladi.
Kalit so'zlar: uglerod oksidi, CO, karboksigemoglobin, toksikologiya, spektrofotometriya, spektroskopiya, gipoksiya, gemoglobin.

Introduction

Carbon (II) oxide (carbon monoxide) is a colorless, odorless, tasteless, fast-acting toxic substance. Very poorly soluble in water, burns with a blue flame:

$$2CO + O_2 = 2CO_2$$

A mixture of carbon monoxide with air in the amount of 16 to 73% can explode. CO, formed as a result of incomplete combustion of organic substances, is widespread in household conditions, industrial environments, and vehicle exhaust gases. Due to its odorlessness and imperceptibility, poisoning with it often occurs suddenly and in most cases is severe. The main toxic property of CO is its strong ability to bind to hemoglobin, which displaces oxygen from hemoglobin and forms stable carboxyhemoglobin. As a result, the tissues of the body and the central nervous system experience severe oxygen deficiency. In forensic medical practice, the determination of carboxyhemoglobin is one of the main criteria for proving CO2 poisoning. This article scientifically analyzes the physical properties of CO, the mechanism of toxic effects, clinical signs, pathological changes, and existing methods for detecting CO.

Toxicological significance. In domestic conditions, carbon monoxide poisoning is a very common phenomenon.

Carbon monoxide is produced when organic substances, coal, gas, and other fuel products are not fully burned. Therefore, staying near stoves and gas stoves for a long time can lead to unpleasant incidents. Carbon monoxide poisoning is

ISSN 3030-3907

most often caused by improper operation of blast furnaces, closing doors and filling sandals with coal before the fire inside the house stoves has completely burned out. On the streets, there are cases of carbon monoxide poisoning caused by car malfunctions.

In the literature, many cases of self-harassment and the harassment of other people by carbon monoxide have also been described. Natural gas fuel used in the national economy can contain 4-30% carbon monoxide, which in turn depends on the type of gas and the method of its production. Therefore, the proper use of gas is also one of the measures to prevent poisoning.

In factories, to obtain high temperatures, a gas mixture called "water gas" is created by injecting steam into glowing coal:

$$C + H_2O = CO + H_2$$

It contains 40-50% carbon monoxide.

It turns out that even during wartime, when shells are fired from artillery weapons and mines explode, many people are poisoned by carbon monoxide - about 30% of carbon monoxide is produced as a result of explosions.

All this indicates the toxicological significance of carbon monoxide. Since carbon monoxide has no odor, poisoning with it occurs quickly and suddenly.

When a person is poisoned by carbon monoxide, they first experience severe headaches, a feeling of weakness, nausea and vomiting, ringing in the ears, a strong heartbeat, weakness, tight jaws, and loss of consciousness due to poisoning and lack of oxygen. Heart activity decreases, breathing slows down - all of this leads to the victim's death. Sometimes carbon monoxide can affect the body extremely quickly, causing convulsions in the victim.

During the pathoanatomical examination of the deceased, the cadaveric spots are dark red, the blood is also dark red due to carboxyhemoglobin, the liver, kidneys, and lungs are also reddened, and there are signs of hemorrhage in the brain tissue.

When a person is poisoned by carbon monoxide, CO easily combines with hemoglobin and forms a stable carboxyhemoglobin compound.

$$HbO_2+CO=HbCO+O_2$$

Hemoglobin loses its ability to transport oxygen in the body, and as a result, the patient dies from oxygen deficiency.

The rate at which carbon monoxide binds to hemoglobin is 300 times greater than the rate at which oxygen binds to hemoglobin. The rate of carbon monoxide release from carboxyhemoglobin is 3600 times slower than the release of oxygen from oxyhemoglobin.

If the carboxyhemoglobin content in the blood is 20-30%, mild poisoning occurs; 30-35% - moderate poisoning; 35-50% - severe poisoning; 50-60% - severe poisoning, seizures, loss of consciousness; when its level reaches 70-80%, death

ISSN 3030-3907

occurs. The changes that occur in the body as a result of such poisoning are presented in Table 1.

In the blood of a person poisoned by carbon monoxide, oxygen and hemoglobin not bound to carbon monoxide - that is, deoxyhemoglobin (Hb), hemoglobin bound to oxygen - oxyhemoglobin (OHb), hemoglobin bound to carbon monoxide - carboxyhemoglobin (COHb), as well as partial methemoglobin (MtHb) may be present.

MtHb does not combine with carbon monoxide. The muscle tissue (meat) of the poisoned person contains deoxymioglobin (MHb), oxymyoglobin (OMHb), and carboxymyoglobin (COMHb).

If carboxyhemoglobin is detected in the blood, it proves the patient's carbon monoxide poisoning.

Table 1

Dependence of carbon monoxide poisoning symptoms on blood carboxyhemoglobin levels.

№	Carboxyhemoglobin content %	Signs of poisoning
1	0-10	No signs of poisoning
2	10-20	Light headache, redness of skin with dilated blood vessels
3	20-30	Sensation of crest on head, temporal pulse
4	30-40	Kukki headache, weakness, dizziness, blurred vision, thirst, vomiting, collapse
5	40-50	Superior state, pronounced collapse, increased respiration and pulse
6	50-60	Further increase in breathing and pulse, periodic recurring convulsions, severe difficulty breathing
7	60-70	In addition to the above condition, slowing of breathing, deterioration of heart function, and death may occur
8	70-80	Extremely weak pulse, severe difficulty breathing, respiratory arrest, and death

Carbon monoxide is excreted from the body only through the respiratory tract. There are different opinions about the storage of carbon monoxide in the human body. According to one of them, carbon monoxide persists in the body for a long time (up to 122 days), and according to another, it cannot be detected after 25-40 hours.

The presence of 0.07-0.08% carbon monoxide in the air of homes and businesses is life-threatening. Its permissible concentration in the air is 0.02 mg/l.

ISSN 3030-3907

Determination of carbon monoxide in cases of poisoning is based on the determination of carboxyhemoglobin in the blood by chemical and physicochemical (spectroscopic, spectrophotometric, photometric, gas chromatographic) methods.

Identification from a biological object. To determine carbon monoxide from the object's composition, the object's blood is separated by crushing and then the blood composition is examined. To determine carbon monoxide in the air, on the contrary, air is passed through the animal's blood, collected in the blood, and then this blood is analyzed.

Carbon monoxide determination by physicochemical methods.

1. Spectroscopic analysis of carbon monoxide. When blood containing carboxyhemoglobin is examined using a spectroscope, Fraunhofer lines (spectra) are visible in the D and E regions of the spectrum. Oxyhemoglobin (OHb) produces an equal spectral absorption of 577-556 nm wavelengths between the D and E regions of the spectrum. The spectra of oxyhemoglobin, deoxymioglobin, and caboxyhemoglobin and their differences are shown in Figure 1.

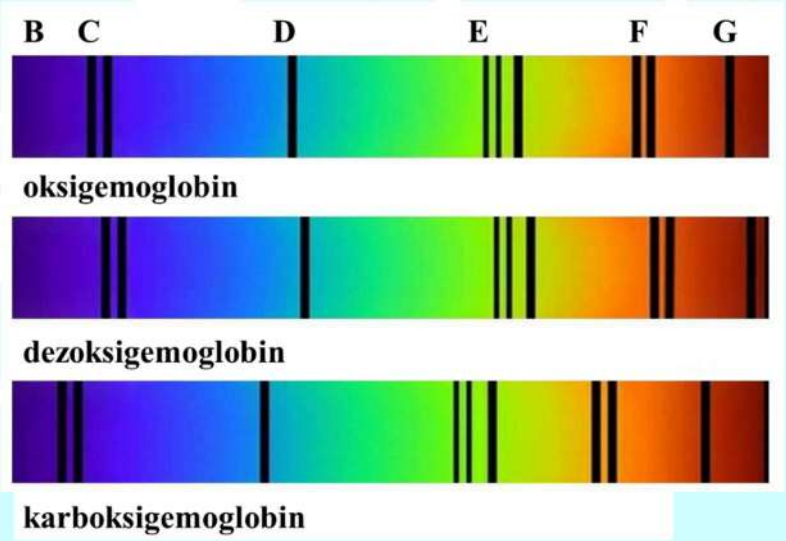


Figure 1. Carbon monoxide poisoning blood spectra.

Carboxyhemoglobin produces spectra in the regions of 564-579 and 523-536 nm. The spectra of oxyhemoglobin under the influence of the reducing agent (ammonium sulfide) combine with each other, forming a wide spectral band in the region of 543-596 nm. Conversely, if carboxyhemoglobin is present in the blood, the spectra in this region remain unchanged. This is because carboxyhemoglobin is not reduced to hemoglobin by ammonium sulfide.

2. Spectrophotometric analysis. The spectrophotometric method is widely used in toxicological chemistry laboratories to detect carbon monoxide and other toxins affecting hemoglobin. The essence of the method is that oxyhemoglobin and carboxyhemoglobin are reduced to deoxyhemoglobin is based on determining.

ISSN 3030-3907

The authenticity and quantity of the above-mentioned hemoglobin-derived substances (deoxyhemoglobin, oxyhemoglobin, and carboxyhemoglobin) can be determined based on their absorption spectra in the 450-620 nm wavelength region. For this, the test blood solution (A), a blood solution consisting of a mixture of carboxyhemoglobin and deoxyhemoglobin (B), and a blood solution containing only carboxyhemoglobin (B) are prepared.

To prepare A-solution, 1 ml of blood without sediments is mixed with 100 ml of phosphate buffer at pH=7.38 or 0.1% aqueous ammonia solution until it reaches 100 ml. Care is taken to ensure the color of the resulting solution is clear. To prepare solution B, solution A is placed in the spectrophotometer cuvette and 3-4 mg of sodium dithionite (hydrosulfate) crystals ($Na_2S_2O_2 \Rightarrow 2H_2O$) are added and thoroughly mixed. In this case, oxyhemoglobin is completely reduced to deoxyhemoglobin. After adding the reagent, the transparency of the solution is controlled.

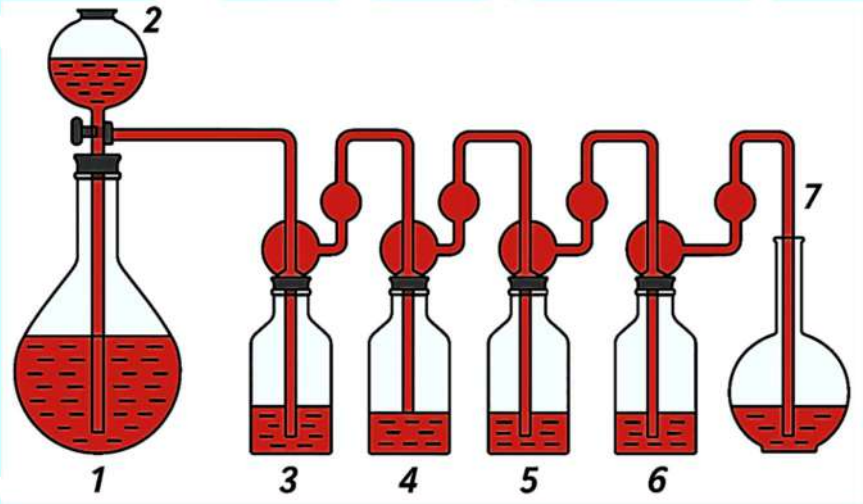


Figure 2. Device for saturating blood with carbon dioxide.

For the preparation of solution B, using a special device (Fig. 2), carbon monoxide formed from concentrated sulfuric acid and formic acid is passed through a 10% alkali solution (3) (containing HCOOH vapors) and then water (4) (purification from NaOH) into a container (5) containing solution A. Here, oxyhemoglobin in the blood completely converts to carboxyhemoglobin. Then, to convert methemoglobin to deoxyhemoglobin, 5-7 mg of sodium dithionite is added to the container and saturated with carbon monoxide again. A solution containing carboxyhemoglobin must also be clear. Using a buffer solution (or corresponding in other variants) or water taken for blood dilution in the second cuvette as a reference solution, the absorption spectra of solutions B and C are measured in the 450-620 nm region, and the indicators of both spectra are plotted on a single diagram. In this case, it is characteristic that deoxyhemoglobin (solution B) forms spectra with one spectral maximum equal to 557 nm, and carboxyhemoglobin forms spectra with two spectral maxima equal to 541 and 571 nm, and in their

ISSN 3030-3907

general scheme, the appearance of intersection points (isobestic) in the regions of 550, 565, and 580 nm is characteristic (Fig. 3).

This diagram is used to determine the quantity. The optical densities of solutions A and B are measured, and the amount of carboxyhemoglobin is determined using the formula. For this, the optical parameters of the tested blood are determined by two monochromatic rays:

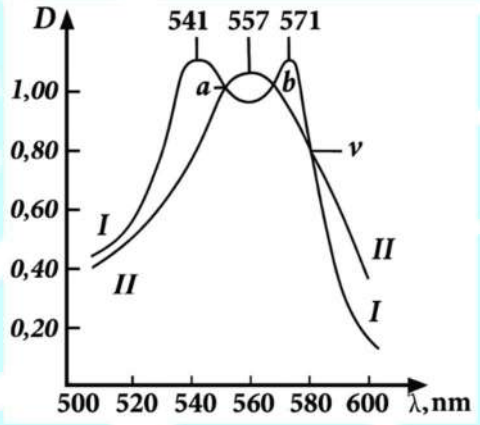


Figure 3. Radiation absorption spectra of I-carboxyhemoglobin, II-deoxyhemoglobin.

It is measured at wavelengths of 534 nm (D1) and 550 nm (D2), since it is at these wavelengths that the distance between the optical parameters of carboxyhemoglobin and deoxyhe-moglobin has the greatest value. From optical indicators, the D2 value is measured at a wavelength of 550 nm at the intersection point of the curves (see the spectral curves of carboxyhemoglobin and deoxyhemoglobin).

The spectral line of kayboxyhemoglobin is determined by saturating blood with carbon monoxide, completely converting it to HbCO3, and then measuring its optical density using a special device.

The spectral line of deoxyhemoglobin is determined by the action of sodium hydrosulfite (reducing agent) on pure blood (not containing carboxyhemoglobin).

Detection procedure. There is also a method for determining carboxyhemoglobin using an ammonia solution instead of a buffer solution. The blood sample is placed in a 100 ml volumetric flask and filled to the mark with a 0.1% aqueous solution of ammonia. After thorough mixing, it is transferred to a 1 cm spectrophotometer cuvette.

Sodium hydrosulfite crystals are added to the solution in the cuvette, the lid is closed, and the optical density of the solution is measured on a spectrophotometer at wavelengths of 534 nm and 550 nm. A 0.1% ammonia solution is used as the reference solution.

Based on the obtained data, the percentage of cyboxyhemoglobin in the blood is calculated using a formula.

ISSN 3030-3907

The percentage of carboxyhemoglobin in the blood is determined by the following formula:

$$P = 100 - \frac{(D_{COHb} - D_{HbCOHb}) * 100}{D_{HbI} * K}$$

Where: P - carboxyhemoglobin content %

DCOHb - optical density of blood in solution V saturated with additional carbon (II) oxide at a wavelength of 538 nm.

Optical density of blood solution B containing DHbCOHb-deoxy and carboxyhemoglobin at 534 nm after the addition of sodium dithionite.

Optical density of D_{HbI}-B solution at 550 nm (isobestic point).

K - coefficient 0.372.Olingan natija is gazi bilan zaharlanishni sud-tibbiy baholashga xizmat qiladi.

Chemical methods. These methods are also based on the fact that carboxyhemoglobin is a more stable substance than oxyhemoglobin.

1) If unpoisoned blood is diluted with water, a brown color appears. If carboxyhemoglobin is present in the blood, it does not change its dark red color.

2) When caustic alkali solutions are applied to normal blood, a bluish-black liquid is formed, while blood poisoned by carbon monoxide retains its color.

3) When a tannin solution is added to normal blood, a gray substance is formed, while blood containing carboxyhemoglobin does not change color.

4) A mixture of yellow blood salt and acetic acid forms a brown color with normal blood; with carbon monoxide poisoning, the blood color does not change.

5) Lead acetate solution stains normal blood a dirty blue color, while blood containing carboxyhemoglobin retains its color.

6) Formalin forms a dirty brown substance with normal blood, and with blood containing carboxyhemoglobin, it does not cause any changes.

7) With a copper sulfate solution, pure blood turns green, and with carbon monoxide poisoning, the blood does not change its dark red color.

In conclusion, carbon monoxide is one of the most widespread toxic gases in household and industrial conditions. Its odorlessness and rapid action increase the risk of carbon monoxide poisoning. The formation of carboxyhemoglobin due to the high affinity of carbon monoxide with hemoglobin can lead to severe hypoxia, central nervous system damage, collapse, seizures, and death. Spectrophotometric and spectroscopic methods for determining the percentage of COHb in the blood are considered the most reliable methods in forensic medical examination. To reduce the risk of carbon monoxide, it is important to improve ventilation systems, properly use fuel, and reduce transport emissions.

ISSN 3030-3907

REFERENCES

1. Ikromov L.T., Tojiyev M.A., Zaynutdinov X.S. Toksikologik kimyodan praktikum. -Toshkent: Fan, 2008.
2. Isroilov I. A., Ro‘ziyev Sh. A. Toksikologiya. - Toshkent: O‘zbekiston tibbiyot nashriyoti, 2018.
3. Nazarov B. T. Umumiy toksikologiya. - Toshkent: Oliy ta’lim nashriyoti, 2015.
4. Yo‘ldoshev A. B. Umumiy toksikologiya asoslari. - Samarqand: SamDU nashriyoti, 2014.
5. Xamidov M. A. Tibbiy kimyo. - Toshkent: Tibbiyot nashriyoti, 2015.
6. Ro‘ziyev S. T. Sud tibbiyotida zaharlanishlar diagnostikasi. - Toshkent: Medpress, 2021.

ISSN 3030-3907