The value of the post mortem analysis of carboxyhemoglobin concentration in the blood – a 15-year study

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SUMMARY
Introduction/Objective The purpose of this retrospective study was to assess carboxyhemoglobin (HbCO) concentration in all autopsy cases of fire and non-fire victims – all those who have been suspected to be under the influence of carbon monoxide (CO) in the Belgrade area during a 15-year period (1990–2005).

Methods Correlations between the concentration of HbCO, circumstances of death, vital signs, and prior health conditions, smoking and history of alcohol-abuse, have been particularly analyzed in 192 autopsy cases.

Results The investigation included 52 (27%) females and 140 (73%) males, their average age being 50.78 years. CO poisoning has been established as the cause of death in 74 cases (38%). The manner of death in 170 cases (89%) was an accident, in eight (3%) suicide, in two (1%) murder, in 11 (6%) natural death, and in one case the manner of death hasn’t been established. Such distribution of the manner of death differentiates this study from others of similar type, and it is conditioned by the specificity of life in this region.

Conclusion The predictors of a high HbCO concentration are the sex, cause of death, manner of death, soot aspiration, lower-degree burns. According to the results, we profiled a typical victim of CO poisoning.

Keywords: carbon monoxide; forensic medicine; epidemiology; HbCO concentration

INTRODUCTION
Carbon monoxide (CO), as one of the most common toxic agents in forensic practice, is a by-product of incomplete combustion of hydrocarbons and it originates mostly from human activity (internal combustion engines, industrial discharges, poorly functioning heating systems).

Exposure to CO can be either intentional or accidental (industry, mines, explosions, exposure to automobile exhaust fumes or in households) [1]. Unintentional fire-related deaths caused by CO poisoning are the most common; however, deaths caused by suicide or homicide are possible, but rare [2].

CO is impossible to detect by a person exposed to it because it is colorless, tasteless, odorless, and non-irritating [3]. When inhaled, CO forms reversible complex with hemoglobin (Hb), known as carboxyhemoglobin (HbCO). Values of HbCO above 50% saturation are usually consistent with death, values 10–50% could indicate that the individual was alive when the fire began, and values below 10% could suggest that the individual had been dead prior to the fire [4].

The goal of this retrospective study was to analyze all autopsy cases in the Belgrade area, suspected to have HbCO, pointing out relations between concentration of HbCO, circumstances of death, vital signs, and prior health condition. We tried to profile the victim of CO poisoning as well as the circumstances of death.

METHODS
A retrospective analysis of 192 autopsy cases (including microscopical and toxicological findings) with suspicion of CO poisoning over a 15-year period (1990–2005) was performed at the Institute of Forensic Medicine, Faculty of Medicine of the University of Belgrade, Serbia. Although CO analysis is not a routine procedure during autopsy, except in arson victims, CO was analyzed when the circumstances of death, the scene investigation, or the autopsy findings (the color of the liver, etc.) indicated CO poisoning. We analyzed concentrations of HbCO in relation to different epidemiological parameters: sex, age, mode, manner and cause of death, circumstances of death, degree of the burns, alcochemistry, soot aspiration, smoking habits, diseases that could have influenced the sensitivity to the toxic effect of CO. For determining the HbCO concentration in the blood taken from the femoral vein, the spectrophotometry method was used (M4 QIII spectrophotometer, Carl Zeiss AG, Oberkochen,
Acquired results are shown in Table 1. Fifty-two females (27%) and 140 males (73%), aged 1–92 years (the average being 50.78 years) were included in the study; males were statistically more frequent than females. There is a statistically significant effect of HbCO concentration depending on the sex (Kruskal–Wallis = 0.589, df = 3, p > 0.05).

There is no statistically significant effect of HbCO concentration depending on the age (p = 0.879).

In 41% of the cases, the concentration of HbCO was up to 10%; in 12% of the cases, it was 11–20%; in 21% of the cases, the concentration was 21–50%; and in 26% of the cases, the concentration was over 50%; the average of the established concentrations of HbCO in the entire sample was 28.09 ± 2.05%.

CO poisoning as the cause of death by itself or in combination with chronic diseases or injuries was established in 74 cases (38%). In 43 cases, the causes of death were burns and their complications, 50 bodies were charred. In 25 cases, violent on non-violent causes of death were established (brain contusion, hemorrhage, complication of mechanical trauma, electrocution, heart infarction).

When observing deaths, assumed to be related to CO poisoning, a balanced distribution regarding the age can be noticed. The highest percentage (36%) of the HbCO concentration of over 50% was in the group of persons up to 20 years of age, while in the other groups the same HbCO concentration was established in 24% of the cases, without a statistically significant difference.

In 76 cases (40%), there were no atherosclerotic changes, while in 116 cases (60%) there was distinct atherosclerosis. There is no statistically significant difference in the HbCO concentration between smokers and non-smokers. Representation of HbCO concentrations in the groups (up to 10%, 11–20%, 21–50%, and over 50%) does not show a statistically significant difference between smokers and non-smokers. Analyzing atherosclerosis in smokers, relating to HbCO; it was found that out of 41 persons with the HbCO concentration of 21–50%, four (10%) had not had atherosclerosis nor had they smoked (χ² = 2.65; df = 3, p = 0.449), nine (22%) had not smoked but had had atherosclerosis, 10 (25%) had smoked but hadn’t had atherosclerosis, and 18 (43%) had been smokers with atherosclerosis (χ² = 6.68; df = 3, p = 0.083, p > 0.05).

The average value of blood alcohol concentration (BAC) was 0.06 ± 0.01 g%; 140 cases (73%) had a concentration of less than 0.03 g%, 42 (22%) had 0.03–0.3 g%, and 10 (5%) had a BAC of more than 0.3 g%. When cross-referenced, HbCO concentrations and BAC did not show a statistically significant effect of degree of inebriation on exposure to CO, and therefore to HbCO (χ² = 8.565, df = 6, p = 0.200, p > 0.05).

In the analyzed sample, there were 22 (11%) psychiatric patients, whereby in this group CO poisoning was the cause of death in 41% of the cases. The link between psychiatric condition and CO poisoning can be seen even if we observe these parameters in the summed sample where the group of psychiatric patients makes up 12% of the causes of death by itself or in combination with chronic diseases or injuries was established (brain contusion, hemorrhage, complication of mechanical trauma, electrocution, heart infarction).

Relating to the scene of event, in 21 cases (11%), the body was found outdoors, and in 171 (89%) cases it was found indoors. Out of 30 victims found in cars, there was...
only one case where the HbCO concentration was over 50%, in 17 cases (61%) an HbCO concentration of up to 10% was established, and in the remaining 36%, the HbCO concentrations were between those two values. Soot aspiration was established in 139 cases (72%), while in 53 cases (28%) it was not found. HbCO was detected in every case with soot aspiration.

By the manner of death, 170 cases (89%) were accidents, eight (3%) were suicides, two (1%) were murders, 11 (6%) were natural deaths, and in one case the cause and the manner of death were not established. Out of the total sample, death was instantaneous in 176 (92%) cases, and in eight cases (4%), survival lasted up to 24 hours, and over 24 hours. When analyzing HbCO concentration relation to the manner of death, it can be noticed that most of the CO poisonings were established in accidental deaths.

Correlations between burn degrees and HbCO concentrations are shown in Table 2. There is a statistically significant correlation between the burn degree and the HbCO concentration – the lower the burn degree, the higher the HbCO concentration ($\chi^2 = 50.67$; df = 15; $p = 0.000$, $p < 0.05$).

### Table 2. Correlation between the degree of burns and carboxyhemoglobin (HbCO) concentrations

<table>
<thead>
<tr>
<th>Burn degree</th>
<th>HbCO concentration (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0–10</td>
</tr>
<tr>
<td>none</td>
<td>1</td>
</tr>
<tr>
<td>I degree</td>
<td>0</td>
</tr>
<tr>
<td>II degree</td>
<td>2</td>
</tr>
<tr>
<td>III degree</td>
<td>16</td>
</tr>
<tr>
<td>Combined II, III, IV</td>
<td>15</td>
</tr>
<tr>
<td>Total</td>
<td>33</td>
</tr>
</tbody>
</table>

Statistical analysis has shown that the predictors of high HbCO concentration are the sex, cause of death, manner of death, soot aspiration, and burn degree.

### DISCUSSION

In the Belgrade area (population of approximately two million), deaths caused by CO poisoning are the second most common among intoxications (drug-related deaths are the first) [5, 6]. The most common source of CO in Belgrade are home, industrial, and traffic fires, as central gas heating appliances are not available. Heat and toxic fumes are the main causes of death during a fire [7]. It is known that with low levels of HbCO in victims distant from a fire rises the possibility of other toxic gases, but as this was a retrospective analysis, other gases (hydrogen cyanide and other methemoglobinemia-causing compounds) were not analyzed during the observed period [8].

The analysis of our sample in relation to the sex shows that males are more represented in the total sample, but higher concentrations of HbCO are more common in females. Being that blood levels of CO are expressed relatively, with Hb saturation, this can be explained with lower absolute quantity of Hb in females alongside with more common anemia, and therefore its quicker saturation rate. Unfortunately, we were not able to compare this to any examples in the literature because we were not able to find any analyses of the type.

Our results coincide with the results of other research – no significant correlation of HbCO concentration and age had been noticed. Although there is no statistically significant difference, the young are more resilient to CO effects, and higher concentrations are necessary for death to occur. This can be explained with the lack of predisposed factors like anemia, cardiovascular, or lung disease.

Our study showed that the analysis of HbCO concentrations was requested and done reasonably. CO poisoning was the cause of death in 38% of the cases (HbCO of over 50%, statistically highly significant), while HbCO as a vital sign was found in 59% of the cases. The most important diagnostic parameter for vitality injury are simultaneous findings of an elevated HbCO concentration (over 10%) and of soot aspiration. There is a statistically highly significant correlation of the HbCO concentration and soot aspiration, unrelated to the scene of event (indoors/outdoors). This is logical, as respiration is necessary for both, but in some cases fire is not accompanied with soot, so the positive HbCO finding is a sufficiently vital sign [5]. In our specimen, all the cases with occurring soot aspiration had a concentration of HbCO higher than 10%, while Bohnert et al. [6] state a 57% correlation. However, soot aspiration was not established in 15 cases where the concentration of HbCO was over 10%. The concentration of HbCO can be elevated even without soot aspiration, which is explained by the presence of HbCO even without a flame, as well as by with the type of the burning material, and by the dynamics of fire development.

The fact that there was a significant correlation between soot aspiration and high HbCO concentration ($\exp B = 11.360$; $p = 0.08$) even with the bodies found outdoors was the result of a quick combustion of large quantities of accelerators on the body (a temperature of up to 1,100°C is reached within seconds). In our conditions, the most common is suicidal self-immolation, although concealed murder must not be ruled out [9, 10, 11].

It was established that in 26 cases (13%), the cause of death was not in any correlation with the effect of high temperature and/or CO poisoning (natural death, brain contusions, etc.) – the bodies were engulfed in flames post-mortally. This confirms the validity of determining the HbCO concentration in all the cases where there is the effect of flame, since even the negative result can contribute to solving the case.

Blood HbCO concentration was statistically highly conditioned by the location where the body had been discovered (indoors/outdoors). Our analysis shows that a high concentration of HbCO is characteristic for bodies found indoors. This can be explained by the fact that, in our conditions, the biggest source of CO is a fire accompanied by oxygen consumption with an inadequate inflow, which leads to CO production. An active person, breathing air containing 5% CO has an HbCO level of about 40% in 30 seconds [7, 12].
However, in our sample, car victims showed completely different results. Namely, in just one case with car victims, the HbCO concentration was high enough to cause death (over 50%), while in other cases (in 36% of the cases the concentration was 11–50%) it indicated the vitality of the injuries sustained before the traffic accident fire. The person was alive for a certain period of time when he/she was engulfed in flames; death was most commonly caused by brain contusion and it did not occur instantly. In closed confines (car cabin), a temperature of about 250°C occurs in five to six minutes, which rapidly causes burns [7]. These results differ from those found in the literature, where it is stated that fire victims dying in closed automobiles uniformly exhibited HbCO levels exceeding 30%, and are conditioned by the fact that suicides by CO from the car exhaust gas were not common in our country, but car accidents followed by fire were common (smuggling gasoline) [9, 13].

Ethanol is a commonly used substance and a high proportion of cases of CO poisoning where ethanol was detected is not surprising [13–16]. Ethanol can potentiate the effects of CO in such a manner that lower levels of HbCO are associated with fatality. In our study, the average value of BAC was 0.06 ± 0.01 g% (the legal cut-off point in our country is 0.03 g%). BAC level in our example does not have a statistically significant effect on the HbCO concentration, which differs from other studies [9, 15]. This could be explained by the specificity of our sample because the analysis was not aimed toward inebriation in the cases of high HbCO concentration – all the cases where HbCO was tested were analyzed regardless of whether it was the cause of death. This study does not support the opinion that ethanol is the key factor in fatal CO intoxication.

Many authors claim that heavy smokers can have HbCO saturation approaching 10% [2, 7]. Unlike in other research, an HbCO concentration of up to 10% is equally present in smokers and non-smokers. One of the possible explanations for these findings could be the fact that in our country there had not been any indoor smoking prohibited until 2010, so non-smokers had been exposed to passive smoking; in addition, our country is among the leading countries in the number of smokers (38% of males, 30% of females according to the Ministry of Health). Persons with coronary artery disease cannot increase coronary blood flow when HbCO saturation is raised acutely [7]. In our study, in the group with atherosclerosis and HbCO ranging 21–50% (13%), CO intoxication could be considered the cause of death. This is even more obvious when atherosclerosis is combined with smoking cigarettes, since these persons died (in our sample) with the HbCO concentration of 21–50% (all other causes of death were excluded). In these cases, the HbCO concentration of over 50% was not established, since atherosclerosis and lung disease caused by smoking had made these individuals more susceptible to the toxic effect of CO in lower concentrations [7].

By analyzing burn degrees and HbCO, we gained statistically reversed correlation between them – the lower the burn degree, the higher the HbCO concentration (expB = -3.31; p = 0.000). This is explained by the fact that persons died from high degree burns before reaching lethal HbCO concentration levels. With charred bodies, death comes as the result of other reasons that come before CO poisoning.

Uniqueness of CO poisoning in our area can be seen from analyzing death by its means, where by far the most common were accidental poisonings, whereas suicidal and homicidal poisonings were very rare, represented with 5%. CO poisoning as a suicide method is almost nonexistent in our country, unlike the USA and Denmark – hence, we did not have enough valid data for a more thorough analysis [13, 17]. One of the characteristics of our sample is the fact that there is a high percentage of persons with psychiatric history in the group of those poisoned by CO (12%), but they mostly died accidentally, which is in agreement with the literature [18].

The present study has several limitations: other toxic gases were not analyzed and representation of suicides and homicides was too low to make more precise conclusions [16, 19].

**CONCLUSION**

A characteristic victim of CO poisoning is an older male person with cardiovascular disease and psychiatric history, a smoker, who died accidentally in an enclosed space from exposure to open flames, with lower degree burns, with aspiration of soot, sober; women die with a higher HbCO concentration. On the other hand, persons with a low concentration of HbCO die of other causes (natural or trauma), mostly carbonized after death.

**REFERENCES**


САЖЕТАК
Увод/Циљ Циљ ретроспективне студије био је одређивање концентрације карбоксихемоглобина (HbCO) у свим случајевима жртава из пожара и оних који нису у вези са њим, на подручју Београда, у периоду 1990–2005, односно свих случајева за које се сумњало да су били изложени угљен-моноксиду (СО).

Методе У 192 случаја обдукције посебно су анализиране: корелација између концентрације HbCO, околности умирања, виталних реакција, претходног здравственог стања, пушења и злоупотребе алкохола.

Резултати Испитивање је обхватало 52 (27%) жене и 140 (73%) мужа просечне старости 50,78 година. Трање CO је утврђено као узрок смрти у 74 случаја (38%). У 170 случајева (89%) радило се о задесима, у 8 (3%) о самоубиствима, у 2 (1%) о убиствима. У 11 случајева (6%) реч је било о природној смрти, док у једном случају узрок смрти није установљен. Оваква расподела диференцира ову студију од сличних и условљена је специфичностима живота у региону.

Закључак Предиктори високе концентрације HbCO су пол, узрок смрти, начин смрти, аспирација чаше и злоупотреба алкохола. На основу резултата направљен је профил жртве тровања CO.

Кључне речи: угљен-моноксид; форензичка медицина; епидемиологија; концентрација HbCO