SECTION 2. CLINICAL MEDICINE

DOI: 10.46299/ISG.2023.MONO.MED.4.2.1

2.1 Modern features of epidemiology, clinical, laboratory and pathohistological diagnosis of narcotic poisoning

The permanent distribution of narcotic and psychotropic substances (hereinafter -NPS) in the world has reached the scale of a global medical and social problem, the solution of which requires international efforts. The constant expansion of the spectrum of NPS complicates the process of their monitoring and risk assessment for public health, as confirmed by Poison Control Centres in the USA, EU countries and many other countries of the world [48-52]. At the same time, according to the Kyiv Toxicology Center, the number of poisonings involving illegal methadone, heroin, buprenorphine, and other opioids, which are often combined by drug users with benzodiazepines, amphetamine, cocaine, various psychotropic substances, and ethanol, has increased significantly in recent years [53-54].

The variety of NPS and their combinations cause the appearance of atypical symptoms in victims, which significantly complicates medical care and causes fatal consequences. The current situation requires a significant strengthening of the capacities of clinical and forensic toxicology laboratories in the implementation of toxicological and analytical screening with the involvement of modern highly efficient methods of chromatography and chromate-mass spectrometry for the identification of new NPS and their biomarkers. Unfortunately, technical and financial limitations allow identification of only a limited range of NPS, which is the reason for the low level of detection and untimely notification of state authorities on the appearance of new NPS with dangerous properties.

Today, in Ukraine, acute poisoning by narcotics is an extremely urgent medical and social problem that is far from being resolved, therefore, there is obviously a need for an in-depth study of the reasons for the increase in fatal poisoning cases in order to prevent them in a timely manner.

For many years, the analysis of cases of fatal poisoning by NPS was carried out with the involvement of epidemiological, clinical, forensic and laboratory methods of research, which was aimed at finding measures to improve the efficiency of the health care system in providing medical assistance for acute household poisoning.

During 2015-2019, the medical data of patients treated with the diagnosis: "Acute narcotic poisoning" (МКХ-10:T40.0-T40.3) in the toxicological center of the Kyiv City Clinical Hospital of Emergency Medical Care (hereinafter - Toxicological Center); statistical data of the State Statistics Service of Ukraine (hereinafter - Derzhkomstat) according to the indicator: "Deaths related to drug use and mortality among drug users" (2018) [55]; statistical data and analytical reports of the European Monitoring Center for Drugs and Drug Addiction (EMCDDA) [56]; statistical data of the Center for Medical Statistics of the Ministry of Health of Ukraine [57]; data from the reports of the Bureau of Forensic Medical Examinations. Research was carried out in accordance with the plan of research work (state registration number - 0112U001133) and in accordance with the Helsinki Declaration of the World Medical Association "Ethical principles of medical research with the participation of a person as an object of research" (1964 p.).

From the mid-1980s to the present day, drugs have turned from isolated and "exotic poisonings" into a dominant group of poisonous substances, the share of which in the total structure of poisonings has reached 42%.

On average, the Toxicological Center provides medical assistance to 2,500 adult patients with acute poisoning of chemical etiology in a hospital, 1,500 patients - on an outpatient basis, round-the-clock telephone consultations and visits of consulting doctors to other hospitals in Kyiv. Toxicological studies on the content of NPS are carried out around the clock, on average; about 25-30 thousand studies of biological material (blood and urine) are performed during the year.

Over the past five years, toxicologists have been studying synthetic and semi-synthetic opioids, psychostimulants, smoking mixtures, sedatives or hypnotics, the diversity of which in terms of chemical composition and specific effects cannot be compared with traditional drugs. In parallel with the emergence of new NPS, the number of poisonings associated with the simultaneous (combined) intake of NPS of various groups, pharmaceuticals and ethanol is increasing.

According to the results of the study of the chemical composition of the urine of patients admitted to the Toxicological Center during 2015-2019, more than 40 groups of substances that led to acute poisonings were found. Drugs of different groups, psychotropic substances used mainly to enhance or prolong the narcotic effect and medicinal products, mainly anti-inflammatory and analgesic, were present in various combinations. Over the past five years, the number of positive tests for opioids has increased 2.3 times, led by illegal home-made drugs: methadone, heroin and morphine. In the structure of psychotropic substances, an increase in the number of positive tests confirming the presence of benzodiazepines, barbiturates, amphetamine, marijuana, and phenothiazine was noted.

In parallel with the changes in the structure of the NPS, the dynamics of the hospital mortality rate (the ratio of the number of deaths from poisoning to the number of those discharged during the year in %) was noted. If the average indicators of hospital mortality in acute poisonings of chemical etiology during the study period were within 3.85±0.65%, then the mortality from NPS poisoning increased by 1.8 times over 5 years. The majority of the dead are men aged 18 to 45.

According to the results of clinical and forensic studies, the main causes of fatal consequences are considered to be: a high dose of poison, the development of complications (post-resuscitation disease, pneumonia, rhabdomyolysis, acute renal failure, etc.), and background concomitant pathology.

It is important to note that the number of deaths from drug poisoning (overdoses) that occurred outside hospitals significantly exceeds the number of deaths in hospital conditions. According to the State Statistics Service of Ukraine for 2014-2018, 1,365 deaths were registered as a result of NPS poisoning. In 2018, 399 cases of deaths related to the use of narcotics and poisoning by them were recorded in Ukraine in 2018, of which 64 (16.04%) cases were related to mental and behavioral disorders due to consumption of narcotics and 335 (98.82%) cases related to external causes of death related to use, namely poisoning or overdoses (Fig. 1).

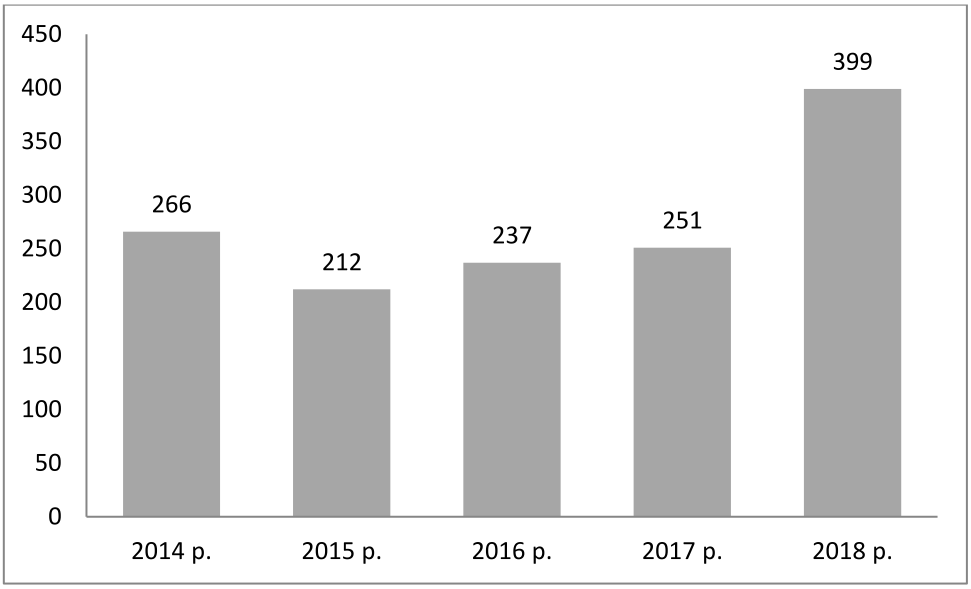

Figure 1. Dynamics of the number of deaths related to poisoning with NPS for 2014-2018 (Source: data of the State Statistics Committee of Ukraine for 2018).

In Ukraine, in 2018, the frequency of deaths related to the use of NPS and poisoning by them, among the population aged 15 to 64, was 10 people for every 1,000,000 population. The breakdown of deaths by gender in 2018 shows that 90.5% were men (361 cases) and 9.5% were women (38 cases). By age group, the largest number of deaths in 2018, namely, 340 cases (85.2%), occurred in the age group of 25-44 years, 13 cases in the age group of 15-24 years, and 46 cases among persons aged 45 and more.

Deaths related to mental and behavioral disorders due to the use of NPS (64 cases) consisted of two causes of death (by ICD-10 codes): 43 cases were related to the abuse of opioids and 21 cases to the abuse of several drugs and other psychotropic substances.

355 cases of death from poisoning by NPS were investigated, it was found that opioid poisoning accounted for 153 cases (codeine and morphine – 54 cases (43.1%), methadone – 76 cases (21.4%), opium – 15 cases (4.2%), synthetic opioids – 8 cases (2.3%)).

144 cases of death from poisoning by other drugs were studied, including 31 (21.5%) cases of poisoning by psychodysleptics (hallucinogens), 2 (1.4%) cases of poisoning by cocaine and 5 (3.5%) cases of poisoning by psychostimulants.

According to state statistics, the number of deaths from drug poisoning in 2018 increased by 1.5 times compared to 2017. In particular, methadone poisoning increased by 2.5 times, and poisoning by unidentified drugs increased almost 3 times.

According to the intention of the action, the cases of death were distributed as follows: 268 cases of death – accidental poisoning; 66 cases of death – poisoning with an unspecified intention; 1 case of death – self-poisoning.

According to the Bureau of Forensic Medical Examinations (hereinafter – Forensic Bureau), the number of deceased persons with psychotropic substances found in their biological material was 427 cases, of which 119 cases were related to opioids, 104 cases – medicinal products containing NPS or precursors, 64 cases - unidentified substances, 56 cases attributed to the simultaneous intake of several NPS, 49 to sedative or hypnotic substances, 20 to stimulants, 11 to hallucinogens, and 2 cases each to the group of cannabinoids and cocaine.

Analyzing the ranges of age groups for which the presence of psychoactive substances in biological material was established in Forensic Bureau, it should be noted that almost 76.1% (325 cases) belonged to the group from 25 to 44 years of the total number of cases (427 cases). Regarding the structure of cases by gender, 84.8% (362 cases) were male, and 15.2% (65 cases) were female.

The share of opioids was 28% of all established facts of the presence of psychotropic substances in biological material, 24.4% accounted for drugs containing NPS or precursors, 15% for substances that were not identified, the rest consisted of other psychoactive substances.

If we compare the data on cases of death based on the biological material to the cases of death associated with their use and poisoning by them, it can be noted that the ranges of age groups coincide ($p \leq 0.05$), that is, in both data samples, more than 75% of all cases belonged to the age group from 25 years to 44 years.

Comparing the data of the Derzhkomstat with the data provided by the Forensic Bureau by gender, we state that more than 80% of individuals in both samples were male. And only in the cross-section of the NPS spectrum, the analyzed data have certain discrepancies ($p>0.1$), but in both data samples, opioids belong to the most common group among NPS.

During 2019, in Ukraine, the results of the implementation of the Strategy of the state policy on drugs for the period until 2020 (hereinafter referred to as the Strategy) were summarized, which determined the directions and mechanisms of reducing the illegal circulation of drugs and ensuring their availability for medical purposes. Thus, with the aim of increasing the availability of pre-medical care for drug addicts and preventing their death in case of opioid overdose, by order of the Ministry of Health of Ukraine dated 15.01.2018 No. 65, registered in the Ministry of Justice of Ukraine on 06.02.2010 under No. 150/31602, the drug "Naloxone" (INN - naloxone hydrochloride) was allowed to be dispensed from pharmacies in the amount of 2 mg without a prescription [58].

In order to improve the provision of first aid to persons in emergency situations, the order of the Ministry of Health of Ukraine dated October 8, 2018 No. 1833, registered in the Ministry of Justice of Ukraine on December 28, 2018 under No. 1500/32952, approved the procedure for providing first aid to persons with suspected opioid overdose, according to which emergency rescuers rescue services, employees of the state fire protection, policemen, pharmaceutical workers, conductors of passenger cars, flight attendants and other persons who do not have a medical education, but according to their official duties must possess practical skills in providing first aid, are allowed to use Naloxone on persons with suspected opioid poisoning [59].

During 2018-2020, based on the results of monitoring the situation with drug trafficking, the Ministry of Health of Ukraine brought the List of narcotic drugs, psychotropic substances and precursors into compliance with international treaties of Ukraine, more than 150 new substances were included in the List. At the same time, discussions continue regarding the prospects of legalizing medical cannabis in Ukraine for medical purposes.

On this issue, the Ministry of Health of Ukraine considers it expedient to provide access to pain-relieving therapy, in particular cannabis-based, to palliative patients.

Also, the Ministry of Health of Ukraine is planning a number of measures that will facilitate patients' access to medical products within the framework of hospice palliative care (this was included in the program of activities of the Ministry of Health of Ukraine for 2021 approved by the Government). In particular, it was planned to include painkillers in the government's reimbursement program and allow the receipt of drugs by electronic prescription.

Also, the Ministry of Health of Ukraine planned to adapt the licensing conditions to EU requirements, in the part that concerns obtaining a license to participate in the reimbursement program. At the same time, it was planned to significantly strengthen state control. In the near future, the Ministry of Health of Ukraine, together with the State Medical Service, plans to approve the application form and instructions for submitting applications for quotas for the production, manufacture, storage, import into and export from the territory of Ukraine of narcotic drugs, psychotropic substances for scientific and medical purposes.

Unfortunately, the regulatory measures planned to be implemented by the Ministry of Health of Ukraine in the near future are unable to prevent the spread of narcotics in Ukraine and the demand for them among young people. The problems of material and financial support of specialized departments and centers that provide emergency medical aid for NPS poisonings and the needs of toxicological laboratories, which should serve hospitals and toxicological laboratories of the Forensic Bureau, remain unresolved.

Thus, the results of epidemiological, clinical, forensic and laboratory studies carried out by various state institutions in the period 2014-2019 testify to the threatening dynamics of an increase in both the number of cases of NPS poisoning and their variety, as a result of which there is a progressive increase in mortality rates, mainly among the male population aged 25-44. It is also important to note that the injection of opioids (methadone, heroin, buprenorphine, etc.) and amphetamine-type stimulants continues to be a feature of NPS use in Ukraine. An increase in the number of deaths associated with the simultaneous use of NPS (methadone, amphetamine, marijuana, benzodiazepines, ethanol, etc.) in various combinations has been noted.

All this characterizes the poisoning of NPS substances as an acute medical and social problem that requires urgent in-depth study in order to minimize the negative impact on the health of the population.

The organization of medical aid for poisoning, in particular drug poisoning, remains a difficult issue, despite its many years of history.

In Ukraine, emergency medical care and clinical toxicology are areas of clinical medicine that today directly deal with issues of diagnosis, treatment, rehabilitation and prevention of poisoning, including those caused by NPS. The clinical manifestation of poisoning (severity of the condition) can range from asymptomatic to such a critical life-threatening condition, depending on the specifics associated with the toxicant, its route and exposure, as well as the availability of timely and comprehensive treatment. Thus, the ability to predict the severity of poisoning and the prognosis for the patient are critical.

In many countries of the world, to speed up the diagnosis of poisonings (including drug overdoses), point scales for the severity of poisonings are used. One of the most common scales is the Poisoning Severity Score (PSS), which was developed in 1990-1994 by the European Association of Poison Centers and Clinical Toxicologists (EAPCCT), the International Program on Chemical Safety (IPCS) and the Commission of the European Union, with the aim of providing a simple and reliable scoring system for describing poisonings and determining their severity [60].

The PSS uses a set of clinical signs and symptoms from 0 to 4, which provides for the registration of the most severe toxic effects, but is not intended to form a prognosis of poisoning. Score 0 - suspected poisoning, I - mild poisoning, II - moderate poisoning, III - severe poisoning, IV - fatal poisoning. Therefore, all patients who died should have received 4 points.

However, in practice this is far from always the case, an example can be modern drugs, substances with delayed effects, etc. In addition to the PSS, other scoring scales of the patient's condition and prognosis in poisonings (NPDS, APACHE IV, SAPS II, TSS) are actively used, which led to a comparison of the sensitivity and specificity of different scales in cases of drug overdoses, as one of the most common types of socially significant poisonings. We summarized modern approaches to the use of standardized point-based scales for assessing the physiological state of a person on the example of international scales: PSS, SAPS II, APACHE IV and the effectiveness of their use in the process of assessing the severity of NPS poisoning. A retrospective evaluation of the medical records of 2,740 patients with narcotic poisoning (opioid syndrome) treated during 2011-2020 at the Toxicological Center was carried out. The data of deceased patients were evaluated separately (a total of 98 people died, hospital mortality was 3.58%), among which 77 cases (78.6%) were associated with the simultaneous consumption of two or more narcotic substances; 34 cases (34.7%) were associated with opioids and alcohol; 21 cases (21.4%) were associated with a combination of opioids, alcohol and psychostimulants.

The following causes of death were established by forensic medical research: toxic hepato-nephropathy – 12 cases (31.57%); syndrome of long-term compression of soft tissues, accompanied by rhabdomyolysis and acute renal failure - 8 (21.05%); acute kidney damage – 5 (13.15%) without rhabdomyolysis; aspiration of gastric contents – 7 (18.42%); community-acquired pneumonia in 6 patients (15.78%). The sensitivity and specificity of the APACHE IV, SAPS II, and PSS scales for assessing the severity of the opioid syndrome and predicting death were evaluated.

The PSS scale was used to assess the degree of severity in 5 positions: 0 - suspected poisoning, I - mild poisoning, II - moderate poisoning, III - severe poisoning, IV - fatal poisoning. The assessment involved the analysis of indicators in 12 groups: gastrointestinal tract, nervous system, circulatory system, respiratory system, metabolism, liver function, kidney, blood, muscular system, local lesions of the eyes, skin, local effects from bites. The degree of severity was determined in points (0-IV).

The APACHE IV scale involved the analysis of the following data: the APS scale (Acute Physiology Score) - assessment of acute physiological changes; GCS scale (Glasgow Coma Scale); age of the patient; accompanying chronic diseases; place of receipt; duration of treatment in the hospital before admission to the intensive care unit (ICU); diagnosis upon admission (116 items); use of emergency surgery; artificial lung ventilation (VLT) in the first 24 hours; oxygenation coefficient $PaO2/FiO2$, etc.

Based on the assessment of the severity of the condition, the risk of a probable fatal outcome in the hospital was calculated. The evaluation of the final result was carried out according to the APACHE IV scale by summation of all indicators, hospital mortality (HM) was calculated according to the formula: HM = x/(x+1)×100%.

The SAPS II scale was used to assess the risk of in-hospital mortality and consisted of the APS block, the laboratory indicators block, the presence of chronic diseases block, indications for ICU admission, and the use of ventilators. The results were interpreted as follows: the minimum value is 0 points, the maximum value is 160 points, the calculation was made according to the formula: logit = (-7.7631) + (0.0737 (SAPS II)) + ((0.9971 (LN ((SAPS II) + 1))), the probability of a fatal outcome = exp (logit) / (1 + (exp (logit)). Data are used during the first 24 hours of the patient's stay in the ICU. At the first stage of the study, a comparative assessment of the severity of the state of poisoning was carried out on various scales (APACHE IV, SAPS II, PSS), after which the correlation indicators between the obtained data were determined (Table 1.).

Table 1.
The degree of correlation of the indicator of the standardized assessment of the severity of poisoning according to the PSS, SAPS II, APACHE IV scales.

Comparison between scales	Pearson's correlation index, r	p ($p \leq 0,05$)
PSS - APACHE IV	-0,016	<0,001
PSS - SAPS II	0,231	0,002
SAPS II -APACHE IV	0,417	0,054

It was found that the severity indicator of the condition in patients with opioid syndrome according to the PSS scale had a moderate negative correlation (r=-0.016, p<0.001) with the data according to the APACHE IV scale, but the assessment according to the SAPS II scale had a statistically significant positive correlation (r=0.231, p=0.002) with data on the PSS scale.

At the second stage of the study, an analysis of the nature of correlations between the indicators of the severity of poisoning according to the PSS, SAPS II, APACHE

IV scales and the actual occurrence of death among patients with opioid syndrome was performed (Table 2).

Table 2.
The degree of correlation of the severity of poisoning according to the PSS, SAPS II, APACHE IV scales and the onset of death among patients with opioid syndrome.

Assessment of fatal prognosis	Pearson's correlation index, r	p (p≤0,05)
APACHE IV, %	0,548	<0,001
SAPS II, %	0,475	0,051
PSS (4 бали)	0,117	0,078

A statistically significant positive correlation was established between the predictive indicator of the occurrence of death according to the APACHE IV scale and its actual occurrence (r=0.548, p<0.001); according to the SAPS II scale, a similar indicator did not have statistical reliability, but approached the confidence interval (r=0.475, p=0.051); no statistically significant relationship was obtained between the assessment of the state of death on the PSS scale and the actual occurrence of death (r=0.117, p=0.078).

The lack of correlation between the severity of poisoning on the PSS and the actual occurrence of death can be explained by the following considerations: PSS includes a large number of end points - clinical and laboratory data from 12 different organ systems and numerous sub- objective variables such as "mild hemolysis", "mild arterial hypotension", and "prolonged cough", reducing inter-rater reliability.

The PSS scale records critical points during the first examination of the patient, however, the dynamic change in the patient's condition during the first 24 hours of stay in the intensive care unit can add significant corrections to the assessment of the severity of poisoning and its prognosis. Thus, if the evaluation is repeated taking into account the changes, the evaluation indicators can be brought closer to the actual ones, but the re-evaluation by the scale is not expected.

PSS includes a large number of variables, many of which are subjective. In comparison, APACHE II includes only 15 variables derived from basic laboratory values and vital signs, and an additional 16 variables that are somewhat subjective (severe organ system dysfunction or or weak immunity). This may partly explain why APACHE II scoring systems are more commonly used than PSS.

However, APACHE II can be used for all conditions, but PSS is only applicable to patients with poisoning. There have been cases where APACHE II has been used in poisoning studies instead of PSS. In addition, the wider introduction of electronic records of medical data and modern statistical tools for extracting and processing these data can make PSS more convenient to use.

Given the features and limitations of the PSS, many researchers and practitioners are skeptical that the scale in its classic form can be used as a reliable research or clinical tool, which is confirmed by the fact that the PSS is extremely rarely used in North America, where the NPDS (National Poison Data System). However, toxicology centers in Europe and other countries use the PSS daily for each poisoning call, which indicates the ease of use of the scale, a similar system is in place in Canada [61].

It should be noted that the NPDS and PSS systems look very similar, with the main difference being that the lack of detail in the NPDS can reduce the reliability of the assessment. The NPDS stratification uses the critical points on which the PSS is built, but the PSS is a more detailed instrument as a result of an extensive testing process and repeated revisions conducted to improve inter-rater consistency of the data.

Today, the process of development and implementation into clinical practice of a new third system for assessing the severity of poisoning, called TSS (Toxic Severity Score) [62], is ongoing. There are also additional organ dysfunction assessment tools, such as the Multi-Organ Dysfunction Scale (MODS) or the Sepsis-Associated Organ Failure Score (SOFA), which aim to determine the trend of overall organ dysfunction in relation to morbidity and mortality.

In the clinical observation of patients, various static and dynamic indicators of the severity of the condition are used. Typically, critical physiological data points are used at a point in time, while dynamic severity scores collect data points over a period of time. Examples of static scores that are commonly used include the Acute and Chronic Physiological Disorders Score (APACHE II) and the Simplified Acute

Severity Score (SAPS). The Estimation of End-Stage Liver Disease (MELD) model is a dynamic estimation, while the Probability of Mortality Model (MDM) can be static if it is run only on admission or dynamic if it is completed every 24 hours. Clinical practice proves that no point assessment of the severity of the patient's condition can be perfect. For the purposes of clinical trials and treatment audits, a standardized way of classifying the severity of poisonings is needed that does not involve re-examination of medical records to judge what happened. Criticisms that apply to the PSS can apply even more to the NPDS system. The proposed TSS items are notable for the fact that they do not include many components that would allow calculation of a PSS or NPDS score and comparison of their new scoring system for the same patients.

All studies using a 5-point poisoning severity scale contribute to the search for new opportunities for collaboration around the world. For example, we could compare the severity of different types of poisoning between countries based on how they regulate the use of chemicals and pesticides, or national consumer protection laws.

Today, the PSS remains the best way to compare the severity of poisoning by different toxic agents, in different groups of patients with reference to a geographic location or a specific country. Taking into account the catastrophic scale of chemical exposure (or chemical stress) that the population of Ukraine suffers today, not only in the war zone and adjacent territories, the value of timely, adequate and cost-effective management of poisonings is undeniable. Thus, in order to adequately assess the severity of poisoning, various standardized scales can be used. However, the assessment of patients with opioid syndrome revealed significant differences in the severity of poisoning and the prognosis of death, which were calculated using different scales. Thus, it was established that the indicator of the severity of the condition in patients with opioid syndrome according to the PSS scale had a moderate negative correlation ($r=-0.016$, $p<0.001$) with the data according to the APACHE IV scale, but the assessment according to the SAPS II scale had a statistically significant positive correlation ($r=0.231$, $p=0.002$) with the PSS scale data. A statistically significant correlation was obtained between the predictive indicator of the occurrence of death according to the APACHE IV scale and its actual occurrence ($r=0.548$, $p<0.001$); on the SAPS II scale ($r=0.475$, $p=0.051$) and on the PSS scale ($r=0.117$, $p=0.078$), no such relationships were established. In addition to the clinical assessment of NPS poisoning, an important aspect of toxicological diagnosis is laboratory confirmation of the presence of a narcotic substance in the patient's biological material. The prevalence of polydrug use and, as a consequence, the increase in the number of overdose cases with the participation of several narcotics, made it possible to establish that there is a significant variability of concentrations of narcotics in the biological material of a corpse in fatal poisonings (overdoses).

Studying the reports of the toxicology departments and the Forensic Bureau, we found that during the last years in Kyiv among drug users, one of the most common combinations is methadone and alcohol [63-66]. However, toxicological screening proves that the biological tissues of a corpse often show non-lethal concentrations of several toxic substances in the absence of signs of injuries and diseases that could lead to death. Under such circumstances, there are difficulties in determining the immediate cause of death, and in formulating the final clinical diagnosis and forensic diagnoses [67, 68].

There is still debate among scientists and professionals about the possibility of death from the combined effects of methadone and alcohol at their non-lethal concentrations. Therefore, the concentrations of methadone, its metabolites and alcohol in the biological material of those who died as a result of combined poisoning were investigated. A retrospective analysis of 275 reports of forensic medical examination of corpses with the results of forensic chemical and forensic histological examination of cases of death associated with methadone and alcohol was carried out. The average interval before conducting a forensic medical examination was 18.3 hours. Blood was available for measurement in all cases, urine in 115 (41.8%) cases.

The sample was formed according to the following criteria: the presence of methadone in the biological tissues of the corpse (blood, urine, bile, and homogenates of the kidney, liver, and brain); the presence of methadone and alcohol in the blood of the corpse; determination by a medical examiner as the cause of death of combined methadone and alcohol poisoning; absence of other injuries, diseases and conditions that could cause death. During the data analysis, the gender, age of the deceased, the circumstances of the discovery of the corpse, the presence of a history of alcohol and drug consumption, the morphological picture of the fatal case (macro- and microscopic data), the results of the forensic chemical examination of the biological tissues of the corpse were taken into account.

The study of biological material for drug content was carried out by the method of liquid chromatography and mass spectrometry (device - Aligent 6850/5973N, column - HP-5MS), the data are presented in fig. 1. To establish the relationship between the concentrations of opioid narcotic substances and alcohol and the probability of death from the combined effect of toxic substances, the Pearson correlation analysis method was used, at $p \leq 0.05$.

It is known that after oral administration, the bioavailability of methadone is 36-100%, and the maximum concentration in plasma is reached after 1.0-7.5 hours. After taking from 10 to 225 mg, the concentration of methadone in the plasma of a healthy person can vary between 65-650 ng/ml, and the maximum concentration is 125-1250 ng/ml. However, it should be taken into account that the concentration of methadone and alcohol in cadaver tissues is influenced by postmortem metabolism, the pre-life state of organs and systems, individual characteristics of the body, the presence of tolerance to narcotic substances and alcohol, taking antiretroviral drugs, etc.

When investigating cases of death due to the toxic effects of methadone and alcohol (the first group, 166 cases), it was established that the average concentration of methadone in the blood corresponded to the level of 850 ng/ml; the average concentration of the main metabolite of methadone (EDDP) - 250 ng/ml; the mean ratio of blood methadone concentration to EDDP was 13.5:1.

In cases of combined poisoning with methadone and alcohol (the second group, 109 cases) with the participation of other psychotropic substances (benzodiazepines, cocaine, amphetamines, cannabinoids, etc.), the concentration of methadone in the blood corresponded to the level of - 275 ng/ml; the average concentration of the main metabolite of methadone (EDDP) in the blood - 120 ng/ml; the mean ratio of blood methadone concentration to blood EDDP was 8.9:1. The average concentration of methadone in urine in the first and second groups corresponded to the level of 520 ng/ml and 450 ng/ml, respectively, and had no statistically significant difference ($p=0.733$). EDDP concentrations also did not differ significantly between experimental groups and amounted to 655 ng/ml versus 900 ng/ml ($p=0.113$). The concentration of methadone in blood, urine, bile and tissue homogenates in both groups had a wide range of indicators (Fig. 2).

Ethanol was present in 101 cases (36.7%) in an average concentration of 0.257‰ (the range of indicators was 0.020-1.740‰). Other prescription drugs that were typically present at therapeutic or residual concentrations were also detected. It was established that with a lethal combination of methadone and alcohol, their minimum concentration in biological tissues was significantly lower (by 2.0-7.5 times) compared to the lethal concentrations determined by the data of scientific and methodical literature in case of poisoning with one narcotic substance of the opioid series or alcohol. Correlation analysis revealed the absence of a statistically significant relationship between the concentration of methadone ($r=0.211$, $p=0.871$) and its metabolites ($r=0.333$, $p=0.125$) in biological tissues, the concentration of alcohol ($r=0.014$, $p=0.782$) in the blood and the probability the onset of death.

In all cases, signs of death with a short agony period were detected (venous full blood of internal organs; widespread and intensively colored corpse spots; presence of hemorrhages under the visceral pleura and under the epicardium; absence of signs of pronounced dystrophy and impaired microcirculation of organs, etc.); on the basis of morphological and forensic chemical studies, other possible causes of violent and non-violent death were excluded.

Thus, according to the results of research, it was established that the combination of methadone and alcohol significantly reduces the lethal concentration of these toxic substances. However, according to the results of the correlation analysis, no statistically significant relationship was found between methadone concentration in biological material, blood alcohol concentration and the probability of death.

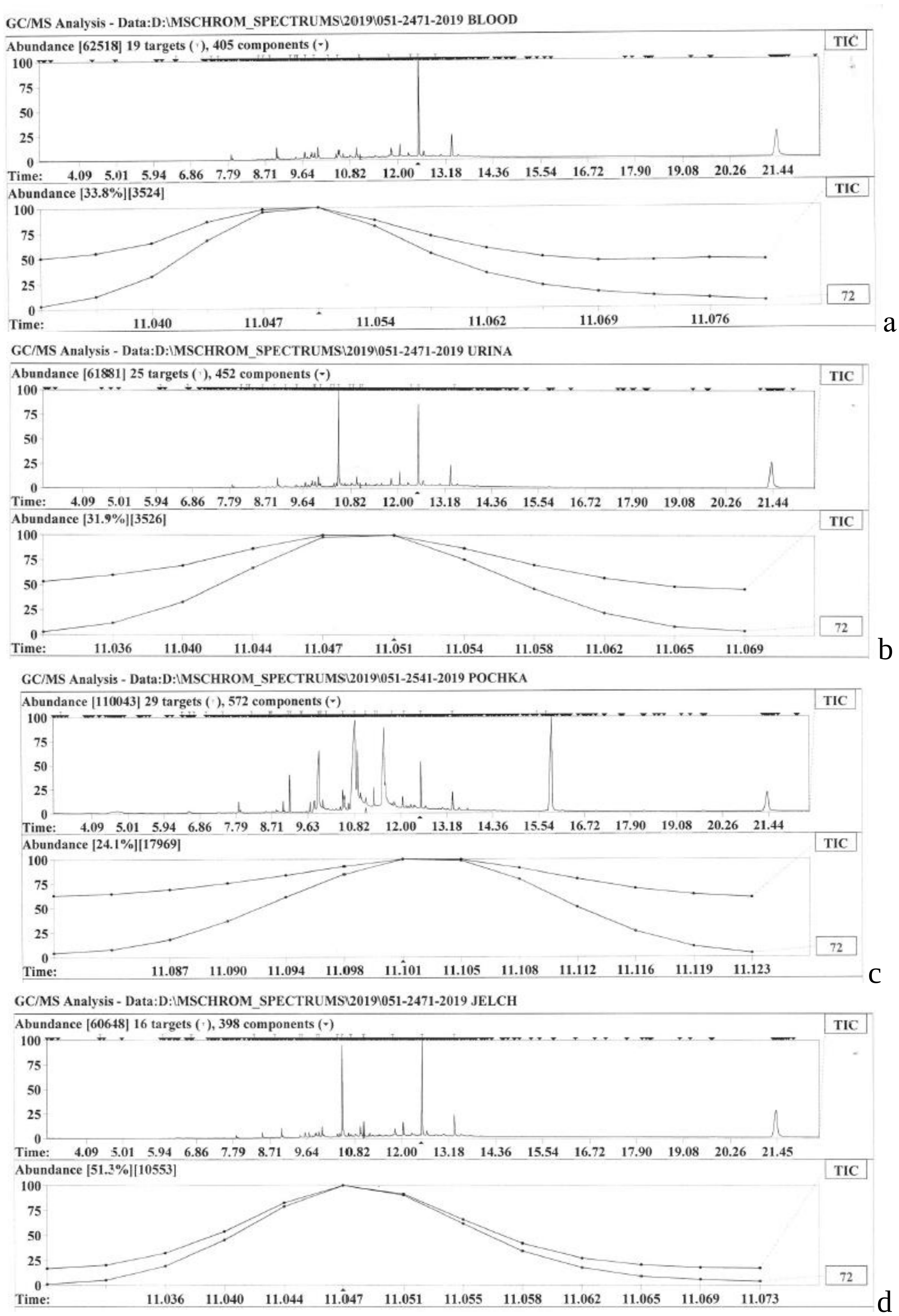

Figure 2. Chromatogram of methadone and its metabolites (EDDP) removed from postmortem samples: blood (a), urine (b), kidney homogenate (c), bile (d). The second phase of elimination. Method of liquid chromatography and mass spectrometry, device - Aligent 6850/5973N, column - HP-5MS.

Illegal use of NPS is accompanied by a high risk of developing acute poisoning due to the lack of control over the doses and composition of the mixtures consumed. In the experiment, with a single intake of a psychoactive substance and alcohol, which exceeds the lethal concentration, a concentration of toxicants is created in biological fluids and tissues, which correlates with the administered dose. However, practice proves that the correlation between concentration and the onset of death is not found in all cases. This can be explained in part by the fact that the redistribution of drugs in the body occurs after death, making it impossible to determine the true concentration of methadone or any other major drug in the period immediately before death.

On the other hand, monitoring drug users on a methadone program is a difficult task. The dose of methadone is calculated on the basis of pharmacokinetic measurements carried out in healthy volunteers who are not drug addicts, so these estimates are imprecise, suggesting a falsely short terminal half-life and a falsely low volume of distribution of methadone in drug addicts, in particular when methadone and alcohol are combined.

It is important to note that the S-isomer of methadone has a small narcotic effect in contrast to the highly effective R-isomer, which is expensive to produce, so a racemic mixture of the R and S forms, or the right and left mirror isomers of methadone, is widely used.

These isomers have different volumes of distribution, which are much larger for the S-form than for the R-form, while the R-form has a shorter half-life and is eliminated much more rapidly than the S-form. At steady state, more of the inactive S-form is found in the blood than the active R-form, and their interaction can probably be used to explain why postmortem methadone concentrations are not significantly different in drug-related deaths, and those whose death is not connected with it.

There was no direct correlation between the concentration of methadone and alcohol and the onset of death in the studied sample, which allows us to assert the existence of other factors of tonatogenesis, in addition to the concentration of methadone and alcohol. An in-depth study of the cause of death in combined drug poisoning requires the involvement of regression models and multivariate analysis.

Thus, it was established that when methadone and alcohol are combined, the minimum lethal concentration in biological tissues is significantly lower than the concentration determined by the scientific literature. The absence of a direct correlation between the concentration of methadone and alcohol (and in some cases other psychotropic substances) in biological tissues and the onset of death was revealed. Reasoning of the cause of death at non-lethal concentrations is possible in the presence of signs of rapid death and the presence of active concentrations of methadone and alcohol, regardless of the presence of non-lethal pathological changes in internal organs.

At the next stage of the research, we studied the causes of fatal cases of NPS poisoning. It is important to note that the level of intraday mortality is one of the main indicators of the effectiveness of diagnosis and treatment in emergency conditions, in particular, in cases of acute poisoning. Predominantly intraday mortality is caused by late seeking medical help, a large dose of poison, the development of life-threatening complications, the presence of concomitant pathology, etc. [69-73]. A retrospective analysis of the medical records of 164 patients who died in the conditions of the Toxicological Center with the diagnosis of "Drug poisoning" for the period 2017-2021 was carried out; the data of sectional and microscopic examinations of tissues (lungs, kidneys, myocardium, brain, liver, spleen) of persons who died outside the hospital as a result of drug poisoning (overdoses) were considered. Tissue pieces fixed in a sufficient amount of formalin were studied. After formalin fixation, alcohol wiring, and paraffin embedding, the sections were stained with hematoxylin and eosin. The preparations were examined using an Olimpus CX 41 microscope in transmitted light, at magnifications of 40, 100, and 400 times.

It was established that the average age of patients was 31.7±7.6 years, men - 149 (90.85%), women - 15 (9.14%). All patients were transported to the hospital by emergency medical teams on the first day after the poisoning. A total of 60 (36.58%) patients died within 24 hours, 104 (63.42%) died within a day or more. Clinical, functional and laboratory tests carried out in the first hours of the patients' stay in the hospital revealed the following pathology: purulent pneumonia - 73 cases (44.5%);

toxic hepato-nephropathy – 23 (13.4%), acute renal failure – 14 (8.5%), long-term soft tissue compression syndrome and rhabdomyolysis – 9 (5.4%), acute cardiopulmonary failure – 19 (11.5%), sepsis - 24 (14.6%).

In 89 (54.2%) cases, according to histopathological studies, changes characteristic of a systemic inflammatory reaction and a chronic inflammatory process (including sepsis) were detected. Signs of microcirculation disorders of varying degrees were observed in all the examined organs: the presence of sludge syndrome; fibrin and mixed thrombi in small vessels; marginal status of lymphocytes and leukocytes; their perivasal location; damage to endotheliocytes; their detachment from the basal membrane; pyknotic changes in the nuclei of endothelial cells.

Phenomena were observed that confirm the increased permeability of the vascular wall and are accompanied by the release of erythrocytes into the perivascular space; hyperplasia of macrophages was observed. During the histological examination of the lungs, purulent endobronchitis and pneumonia were revealed: a large number of hemosiderophages were observed in the space of expanded alveoli; isolated clusters of exfoliated bronchial epithelium; hyaline membranes were found in the alveoli in separate fields of view; sharp congestion of microcirculatory vessels, single mixed thrombi in their lumen, small focal hemorrhages.

In cases of rapid (up to 6 hours from the moment of hospitalization) death from a narcotic overdose in the lungs, full blood, hemorrhages in the alveoli, swelling of the interalveolar membranes, foamy edematous fluid in the alveoli were observed; desquamation of alveolocytes, obstruction of the openings of small bronchi and bronchioles by desquamated epithelium of the mucous membrane.

During the examination of the brain, the following changes were determined: a moderate degree of pronounced perivascular and pericellular edema, sharp congestion of the vessels of the microcirculatory channel with the presence of a sludge phenomenon and single fibrin and mixed thrombi in small vessels; lympho-leukocyte infiltration of the vascular wall with perivasal arrangement of erythrocytes and few leukocytes was observed in separate fields of vision; small foci of hemorrhage in brain tissue; dystrophy of individual neurons; swelling and congestion of the vessels of the medulla oblongata. Dystrophic changes in the endothelium, disorganization of connective tissue and sclerosis of their walls were observed in the vessels of the brain. Changes in neurons demonstrated dystrophy: chromatolysis, karyo- and cytolysis, disintegration of processes, followed by their zonal disappearance.

During the examination of the kidneys, the following changes were revealed: sharply expressed pleurisy of the cerebral and cortical layers; glomeruli with moderate fullness of capillary loops; proximal and distal tubules with signs of granular dystrophy of the epithelium; edema of the epithelium of vessels of the microcirculatory channel; signs of stasis and sludge phenomenon.

During the examination of the myocardium, there was a sharp fullness of blood vessels (erythrocyte stasis, sludge phenomenon), marginal standing of lymphocytes and single leukocytes with a perivasal location; chronic diffuse, mainly lymphocytic myocarditis. The results of the study indicate the presence of clinical signs of endocarditis. Marked dystrophic and sclerotic changes were observed in the myocardium, perivascular cardiosclerosis, lysis, atrophy, metabolic and contractile lesions of cardiomyocytes followed by cardiosclerosis, and lymphohistiocytic infiltration occurred in the walls of the vessels of the microcirculatory channel.

In all cases, portal mononuclear hepatitis with a significant content of lymphocytes and macrophages in the infiltrates, or with the formation of lympho-macrophagic granulomas in the liver parenchyma, sometimes with the appearance of germinal centers, was detected in the liver in all cases. The peculiarity of such hepatitis is that it has no signs of fibrosis of the portal tracts.

When examining the spleen, the following was observed: the red pulp is full of blood; hyperplasia of a part of lymphoid follicles with the formation of large germinative centers with a macrophage reaction inside.

In general, there was marked hyperplasia of lymphoid follicles with the formation of large germinal centers with a macrophage reaction, which is a sign of antigenic stimulation and a morphological substrate of humoral immunity. In the germinal centers, karyorrhexis and phagocytosis by macrophages of nuclear fragments of dead lymphocytes was observed.

It should be noted that the clinical and pathogistological manifestations of the toxic process usually do not depend on the group of drugs, but in most cases are determined by the duration of the drug-related illness, which subsequently forms various multiorgan lesions. The results of complex epidemiological, clinical, chemical-analytical and pathogistological studies demonstrate that modern drug poisoning (overdose) is an extremely dangerous type of poisoning, which is confirmed by high rates of prevalence and daily mortality.

The presence in drug users of a variety of concomitant pathologies of many organs and systems (lungs, heart, liver, kidneys, etc.) increases the toxic effect of narcotic substances and causes the rapid development of complications, which leads to death even with non-lethal concentrations of narcotic drugs found postmortem in blood and urine and tissue homogenates (brain, kidney, liver, bile).

Further monitoring of NPSs and studies of clinic-pathological parallels in fatal poisonings will allow us to expand our understanding of the mechanisms of toxicity of new NPSs and their combined effects.