

Clinical Symptoms of the Systemic Inflammatory Response in Acute Severe Poisoning by Agents Affecting the Central Nervous System

Kuvatov Zoxid Xaitovich

Bukhara State Medical Institute.

Relevance. Currently, acute poisoning with substances affecting the central nervous system in the general structure of exogenous intoxications accounts for up to 50-60% of cases, of which up to 25% are severe and extremely severe lesions leading to the formation of critical conditions and requiring intensive care (Bonitenko E.Yu. et al., 2018; Luzhnikov E.A. et al., 2016; Bohnert A.S. et al., 2019; Muller D. et al., 2017; Taheri F. et al., 2015). The causes of widespread acute poisoning include the uncontrolled use of various medicines in everyday life, suicidal attempts, as well as the use of narcotic drugs for the purpose of drug intoxication (Rokhlina M.L., 2019; Nordstrom D. L. et al., 2017; Taheri F. et al., 2018). The severity of the condition of patients with acute severe poisoning by drugs affecting the central nervous system is due to pronounced disorders of the functions of various organs and body systems due to the specific action of xenobiotics (Luzhnikov E.A. et al., 2016), as well as developing hypoxia (Livanov G.A. et al., 2014; Hashemian M. et al., 2016), leading to the development of life-threatening complications, which often determines the outcome of chemical trauma. A number of authors note that hypoxia in critical conditions causes the formation of a systemic inflammatory reaction consisting in increased production of proinflammatory mediators, activation of cytokines and kinins, increased vascular permeability, increased blood viscosity and microthrombosis (Chereshnev V.A. et al., 2019; Cavailon J.M. et al., 2016; Ramakrishnan S. et al., 2019). Systemic inflammatory reaction and hypoxia are always associated with activation of proteolytic processes, coagulation and fibrinolytic systems (Sanotsky V.I., 2013; Alekhovich A.V., 2020). Numerous authors have shown in their works that with intensive and prolonged action of inflammatory factors, numerous disorders develop both at the cellular and organ levels (Gusev E.Yu. et al., 2018; Saveliev V.S. et al., 2017; Alberti C. et al., 2015). There are reports in the literature on widely used diagnostic algorithms for systemic inflammatory response syndrome in patients with intensive care, therapeutic, and surgical profiles, which allows early diagnosis of infectious complications and timely initiation of therapy (Zhevlakova Yu.A., 2017; Akimova V.N. et al., 2016; Ratzinger F. et al., 2015; Boehme A. K. et al., 2014). However, the possibility of their use in clinical toxicology has not been considered and investigated to date. There are no generalizing comprehensive studies on the problem of systemic inflammation in acute poisoning by drugs affecting the central nervous system, and methods for correcting this component of the pathogenesis of acute exotoxicosis are insufficiently defined. The mechanisms of pharmacological effects on pathological reactions that are components of the systemic inflammatory reaction syndrome, in particular such as proteolysis and the blood clotting system, have not been determined, which seems relevant [1.3.5.7.9.11.13].

There is no information about the role of systemic inflammatory reaction syndrome in the development of infectious complications in patients with acute poisoning by drugs affecting the central nervous system, which are one of the reasons for the increase in the duration of treatment of patients and possible risks of death in the somatogenic phase of acute poisoning.

The purpose of the work. To identify the importance of immune system dysfunction in the pathogenesis of inflammatory complications in acute poisoning by drugs affecting the central nervous system and to identify ways of correction in order to improve the effectiveness of basic detoxification therapy.

Research methods. Patients with severe acute poisoning, with a level of depression of consciousness on the Glasgow coma scale of 7 points or lower, which corresponds to superficial and deep comas complicated by respiratory disorders and central hemodynamics, will be selected for the study. General clinical laboratory tests will include a general blood test with a detailed leukocyte formula and a biochemical analysis with the determination of glucose, lactate, total protein, electrolytes (K, Na, Cl), bilirubin with fractions, urea, creatinine, amylase, ALT, AsT, LDH, coagulogram. All patients will undergo a comprehensive examination, including X-ray examination of the chest organs, ultrasound examination of the abdominal cavity, electrocoagulography, electrocardiography, chemical and toxicological examination of urine, biochemical, hematological, immunological laboratory tests.

Immunological studies will include determination of the concentration of cytokines TNF α , IL-1 β , IFN- γ IL-10 with a set of reagents (Vector-Best, Russia) by solid-phase enzyme immunoassay (ELISA). To determine the indicators taken as a physiological norm, 20 healthy donors will be examined.

The phagocytic activity of pseudoeosinophils in broiler chickens was evaluated according to the method of L.Z.Klechikov (1967), while the phagocytic number (PH) and phagocytic index (PHI) were determined. To do this, a suspension of latex particles in the size of 1-1.3.5 microns was prepared. The initial latex suspension was washed three times with saline sodium chloride solution by centrifugation at 3000 rpm for 10 minutes. The precipitate was resuspended in medium 199, the number of particles was counted in the Goryaev chamber and brought to a concentration of 100,000 in 1 μ l. Then 0.2 ml of the prepared latex suspension was added to 0.2 ml of blood. Incubation was carried out in a thermostat at 37 ° C for 30 minutes with regular shaking. Then 1 drop of 3% glutaraldehyde was added to the tubes and centrifuged at 1000 rpm for 5 minutes. The filler liquid was removed, the precipitate was suspended in 0.1 ml of distilled water and smears were prepared. The smears were fixed with methanol and stained with azur-eosin. The data obtained were recorded under an immersion microscope system. 100 cells were counted and the PH and PHI were determined. Test of spontaneous and latex-induced reduction of nitrosine tetrazolium (HCT test) they were placed in a volume of 0.2 ml in centrifuge tubes. To 0.2 ml of blood placed in centrifuge tubes, 0.2 ml of phosphate-salt buffer solution (FSB) and 0.04 ml of HCT were added to the first; 0.2 ml of prepared latex suspension and 0.04 ml of HCT were added to the second. The mixtures in test tubes were incubated for 30 minutes at a temperature of 37 ° C, shaking regularly.

Then centrifuged at 1000 rpm for 5 minutes. The filler fluid was drained, suspended in 0.1 ml of FSB and smears were prepared. The analysis of spontaneous (in the first tube) and latex-induced (in the second tube) reduction of HCT was carried out under an immersion microscope system by counting the number of activated pseudoeosinophils containing dark violet grains of formazane as a percentage (ShopFR.F., Mattar J.et al., 1984). 63 Identification of T lymphocytes was carried out by the formation of rosettes with sheep erythrocytes (E-ROCK) (Wybran et al., 1972; Jondal, 1984) and lymphocytes for the detection of receptors for the third component of complement (EAC sockets) (E.N. Mendes, 1973). The bactericidal, lysozyme and complementary activity of blood serum was determined by the method described by P.A. Emelianenko et al.

The level of immunoglobulins in the blood serum of animals was determined by simple radial immunodiffusion using monospecific antisera.

The activity of β -lysines was determined by the method of O.V. Bukharin et al. (1972). The method is based on the ability of β -lysines to lyse *Bacillus subtilis*. Circulating immune complexes were determined by the method of Y.A. Grinevich and N.I. Alferov. The method is based on the selective precipitation of antigen-antibody complexes in a 3.75% solution of polyethylene glycol with a molecular weight of 6000, followed by photometric determination of the density of the precipitate. Lysosomal cationic proteins in broiler chickens were determined by V.N. Chebotkevich and S.I. Lyutinsky (1998). Biochemical blood tests were performed using a semi-automatic biochemical blood analyzer StatFax1904+ (AwarenessTechnologyInc.) using standardized VitalDiagnosticsSpb reagents and the following conditions for setting reactions: total

bilirubin by the Andrasik-Grof method (incubation for 20 minutes at a temperature of 20 °C, $\lambda=545$ nm, optical path length 10 mm, lower limit of linearity 8 mmol/l, the upper limit of linearity is 410 mmol/l in comparison with an empty and calibrated bilirubin sample of 171 mmol/l); glucose by enzymatic colorimetric method (incubation - 10 minutes at 37 °C, $\lambda = 505$ nm, optical path length - 10 mm, lower limit of linearity - 1 mmol /l, upper limit of linearity - 30 mmol/l compared to 64 with an empty and calibrated glucose sample - 10 mmol / l); total protein - by biuretic method (incubation - 30 minutes at a temperature of 20 °C, $\lambda= 545$ nm, optical path length - 10 mm, lower limit of linearity - 0 g/l, upper limit of linearity - 120 g/l, compared with a blank and calibrated blood protein sample - 70 g/l); Alanine aminotransferase and aspartate aminotransferase - by enzymatic kinetic method (without incubation at a reaction mixture temperature of 37 °C, $\lambda=340$ nm, optical path length - 10 mm, lag phase - 90 s, reading time - 60 s, factor - 1746, against a blank sample); cholesterol - by enzymatic colorimetric method (incubation - 5 min. at a temperature of 20 °C, $\lambda = 500$ nm, the length of the optical path is 10 mm, the lower limit of linearity is 0.5 mmol/l, the upper limit of linearity is 25.8 mmol/ l, compared with a blank and calibrated sample of serum cholesterol -5.17 mmol /l); triglycerides - by enzymatic colorimetric method (incubation - 5 minutes at 37 °C, $\lambda= 505$ nm, optical path length - 10 mm, lower limit of linearity - 0.0 mg/dl, upper limit of linearity - 700.0 mg/dl, in comparison with a blank and calibrated sample of serum cholesterol - 250.0 mg/dldl); albumin is a method for determining the concentration of albumin in blood serum by reaction with bromocresol green (without incubation at a reaction mixture temperature of 37 °C, $\lambda = 630$ nm, optical path length is 10 mm, the lower limit of linearity is 20 g/ l, the upper limit of linearity is 60 g/ l, compared with idle and calibration sample of albumin in blood serum - 60 g/l); creatinine is a pseudokinetic two-point method according to the Jaffe reaction (without incubation, at a reaction mixture temperature of 37 °C, $\lambda=505$ nm, optical path length - 10 mm, lag phase - 30 s, reading time - 60 s, factor = 2154, lower limit of linearity - 0 mmol/l, upper limit linearity - 1000 mmol/l, compared with a blank and calibration sample of creatinine in blood serum - 177 mmol/l); urea by urease method (without incubation, at a reaction mixture temperature of 37 °C, $\lambda=340$ nm, optical path length - 10 mm, lag phase - 60 s, reading time - 60 s, lower limit of linearity - 0 mmol/l, 65 upper limit of linearity - 200 mmol/l, in comparison with a blank and calibration sample of creatinine in blood serum - 80 mmol/l). Calibration and verification of the biochemical analyzer was carried out within the framework of in-laboratory control with the formulation of studies of standardized samples of artificial blood serum with normative and pathological values of indicators produced by Vital Diagnostics Spb. The study of the clinical condition of cattle was carried out using generally accepted methods [2.4.6.8.10.12].

The indices of the fabricium sac and thymus of broiler chickens were calculated by the formula: $I = m / M * 100$, where - m is the mass of the organ, g; M is the body weight, g. The development of internal organs was determined by opening chickens in the conditions of the Department of Morphology, Pathology, Pharmacy and Non-communicable Diseases of the Bashkir State Agrarian University with further assessment the mass and size of the internal organs. Anatomical and morphological method was used to study the shape, size and topography of organs. The material of the study was the central and peripheral organs of immunity and myocardium. The internal organs were weighed on an analytical balance VLKT-500 with an accuracy of up to 30 mg. Based on the results obtained, the relative mass of the organ was calculated. Linear parameters were measured using a vernier caliper and a b1mm stage ruler. During histological and histochemical studies, the tissues were fixed in a 10% solution of neutral formalin. Next, the hearts of rats of all the studied groups were cut across into 3 sectors of the same thickness: the upper (base of the heart), the middle and the lower (tip of the heart). Tissue samples were dehydrated in a series of alcohols of increasing concentration and poured into paraffin according to a generally accepted method. The sections were prepared on a LEICA RM 2145 microtome (Germany), which were stained with hematoxylin and eosin, according to Mallory. 66 The study and visualization of the preparations were carried out using a Leica DMD 108 light microscope (Germany) with specialized software for setting management and image

capture. During immunohistochemical studies, paraffin sections with a thickness of 4 microns were stained using the Leica Microsystems Bond™ immunohistostainer (Germany). CD 68, PCNA, CASP3, Timp 2 in a 1:300 ratio were used as the first antibodies (Santa Cruz Biotechnology, USA). The indirect streptavidin-biotin detection system LeicaBOND (Novocastra™, Germany) was used for unmasking. The specificity of the reaction was assessed by staining sections without the first antibodies. The study and visualization of the preparations were carried out using a Leica DMD 108 light microscope (Germany) with specialized software for control of settings and image capture and an AxioImager Z1 microscope equipped with a ProgRes C3 photo nozzle and an Axiovision image analysis program (C. Zeiss, Germany) with multiple magnifications of the lens and eyepiece 4, 10, 20, 40.63 (oil), 100 (oil), 400 (oil).

Conclusion. The analysis of morphometric data was carried out using the method of two-factor analysis of variance and the use of nonparametric methods-single-factor analysis of variance by Kraskel-Wallace and comparison of uncorrelated data by the Mann-Whitney method in the Statistica7.0 program. During the experimental part of experiments on laboratory animals, an adrenaline model of myocardial injury was used. This model of myocardial damage is similar in nature to stress damage to the heart, which occurs in sports horses when physical exertion and functional abilities of the heart do not match during training and competitions.

LIST OF LITERATURE

1. Авзалов, Р.Х. Влияние биологически активных веществ (БАВ) на гематологические показатели и бактерицидную активность сыворотки крови кур // Пути повышения эффективности АПК в условиях вступления России в ВТО. Материалы Международной научно-практической конференции. – Оренбург, 2003. - С. 216-218.
2. Авзалов, Р.Х. Влияние различных режимов применения Селенита натрия на гематологический и иммунный статус и прирост живой массы цыплят кросса «Смена» // Пути повышения эффективности АПК в условиях вступления России в ВТО. Материалы Международной научно-практической конференции. – Оренбург, 2003. - С. 219-220.
3. Базекин, Г.В. Научно-экспериментальное обоснование применения глицирризиновой кислоты и её производных в ветеринарии / Г.В. Базекин, А.Ф. Исмагилова. – Уфа: БГАУ, 2010. - 86 с.
4. Базекин, Г.В. Влияние глицирризиновой кислоты на ветеринарносанитарные показатели молока во время хронической интоксикации фосфорорганическими соединениями / Г. В. Базекин, А. Ф. Исмагилова // Проблемы ветеринарной санитарии, гигиены и экологии. - 2012. - № 1. - С. 231-237.
5. Базекин, Г.В. Изучение токсикологических свойств лекарственных средств на основе новых производных глицирризиновой кислоты / Г.В. Базекин, А.Ф. Исмагилова // Ученые записки Казанской 279 государственной академии ветеринарной медицины им. Н.Э. Баумана. - 2012. - Т. 210. - С. 20-23.
6. Гадиев, Р.Р. Эффективность использования Бетулина при выращивании цыплят-бройлеров / Р.Р. Гадиев // Российский электронный научный журнал. - 2015. - С.89-96.
7. Гадиев, Р.Р. Эффективность использования биологически активных добавок в рационах цыплят-бройлеров и кур-несушек: монография / Р.Р. Гадиев. – Кинель: РИО СГСХА, 2017. – 208 с.
8. Ермаков, С.Е. Обмен веществ и продуктивные качества цыплятбройлеров при включении в их рацион энергопротеинового концентрата «ЭСПК»: специальность «Физиология»: автореф. дис. ...канд. биол наук / Ермаков Сергей Евгеньевич; ФГБОУ ВО «Брянский гос. аграрный ун-т. - Брянск, 2017. – 20 с. - Место защиты: ФГБОУ ВО «Белгородский гос. аграрный ун-т имени В.Я. Горина».

9. Исмагилова, А. Ф. Влияние глицирризиновой кислоты на рост, развитие, естественную резистентность и антиоксидантный статус больных острой формой бронхопневмонии телят / А. Ф. Исмагилова, Г. В. Базекин // Ветеринарный врач. - 2015. -№ 3. - С. 25-28.
10. Кузнецова, А.В. Продуктивность гусей при использовании кормовой добавки Ветосел Е форте: специальность «Частная зоотехния, технология производства продуктов животноводства кормление сельскохозяйственных животных и технология корм» 06.02.10: автореф. дис. ... канд. с.-х. наук / Кузнецова Анна Васильевна; ФГБОУ ВПО «Курганская гос. сельскохозяйственная академия имени Т.С. Мальцева». – Курган, 2016. – 19 с.
11. Кулаченко, И.В. Развитие и функциональное состояние иммунокомпетентных и детоксикационных органов цыплят-бройлеров при воздействии липосомных фитопрепаратов // Актуальные вопросы сельскохозяйственной биологии. - 2017. - С.13-20.
12. Родькина, А.А. Некроз и апоптоз в фабрициевой сумке у цыплят при болезни Гамборо /А.А. Родькина // Матер. Всерос. научно-метод. конф. патологоанатомов ветеринарной медицины. - Москва, 2003. - С.117-118. 179.
13. Сабетова, К.Д. Влияние препарата «Кудесан» на клиническую картину у телят при миокардиодистрофии / К. Д. Сабетова // Актуальные проблемы науки в агропромышленном комплексе: материалы 69-й Международной научно-практ. конф. в 3-х томах; под ред. Ю. В. Панкратова, Н.Ю. Парамоновой. - Караваево: Костромская государственная сельскохозяйственная академия, 2018, - С. 219-223.