

**TRANSFORMATION PRODUCTS AND MICROBIOLOGICAL ACTIVITY OF ANTIBACTERIAL DRUGS ASSESSED IN ANIMAL MODELS**

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**ABSTRACT**

Transformed drugs can exhibit a wide range of new toxicological effects compared to those of their original compounds. Conventional wastewater treatment technologies are only able to partially remove many pharmaceutical preparations and their metabolites from the environment, if at all, resulting in their accumulation in soils and agricultural products. These compounds can then enter the human body via the food chain, which contributes greater selection for resistance of microorganisms to these drugs. In this study, we investigated the biotransformation of ciprofloxacin, metronidazole, and adduct iodine in rats. The sensitivity *Staphylococcus aureus*, *Enterococcus faecalis*, and *Escherichia coli* strains to the used drugs was then determined *in vitro* and *in vivo* in a mouse model of sepsis. Ciprofloxacin, metronidazole, and iodine adduct biotransformation products were all detected in the urine and feces of rats during the 7-day course of application. The highest concentrations of ciprofloxacin and metronidazole were found in the urine, whereas the highest concentration of iodides was detected in the feces. *In vitro* studies showed that all three strains were sensitive to the test drugs. Treatment of mice with metronidazole, ciprofloxacin, and iodine adduct caused positive dynamics of the septicopyemia course, but resulted in the development of resistance to metronidazole. These results highlight the necessity to include detection of biotransformed pharmaceutical preparations and their metabolites in open water sources as part of routine monitoring programs of the Republic of Kazakhstan.

**Key words:** pharmaceutical preparations, ciprofloxacin, metronidazole, iodine adducts, biotransformation, environment, resistance, test strains of microorganisms, laboratory animals, sepsis, morphostructure

**INTRODUCTION**

One route of pharmaceuticals entry into environment is the flow of treated wastewater and sludge to agricultural areas or open water sources. In the environment, their further transformation – biotransformation in living organisms and under abiotic factors influence takes place, for example, phototransformation, thermotransformation, etc. [1-3]. Pharmaceutical compounds continuously released into the wastewater via sewer system, then pass through treatment plant and reach soil and water

in an unmodified form or as transformed products [4]. More than 3.500 pharmaceutical compounds, excluding metabolites and transformation products, are found in surface waters. Nonsteroidal anti-inflammatory drugs (NSAIDs) are the most frequently reported [5].

After receiving pharmacological agents in humans and animals are metabolized by group of enzymes, synthesized mainly by the liver and intestines. These enzymes have wide specificity and are able to form various metabolic products. It is accepted to distinguish 3 phases of drug structural

modification. Thus, in phase I, key enzyme is cytochrome P450 (CYP), which oxidizes substrate by joining reactive and polar functional hydroxyl group or oxygen atom. Depending on the substrate, CYP can produce highly active derivative, which can modify other molecules in the cell, including macromolecules, with the development of cytotoxicity risk. In phase II, conjugation of activated chemical with charged components occurs for effective export from the cell, molecular weight of substance also increases and its reactivity decreases. In phase III, conjugated chemical removed from the cell into extracellular medium [6].

Pharmacological agents are characterized by excretion of drugs in unchanged form or in the form of metabolites. One approach for detection of endogenously formed transformation products is their identification in blood, urine and feces.

One social health problem is the development of antibiotic resistance in bacteria. This is associated with their uncontrolled use, resulting in the treatment of infections is not effective [7-9]. It is shown that only small part of antibiotics undergoes complete biodegradation, and basic part of them enters environment in unchanged or in form of metabolites [10].

Total and uncontrolled use of antibiotics can lead to sub-inhibitory concentrations (sub-MICs) in the environment. Sub-MICs can increase genetic and phenotypic variability of bacteria, affect signal transfer from cells to cells, gene expression in bacterial populations. These effects can influence competitive interactions, influence the structure and functioning of microbial communities [11].

Main cause of antibiotic resistance is horizontal gene transfer: conjugative plasmids and transposons encode antibiotic resistance and exactly they are exchanged between bacterial species. Environmental factors, including species interactions and the level of spatial structuring of habitats, affect the structure of the bacterial community and horizontal transmission of antibiotic resistance genes by maintaining high bacterial density and metabolic activity [12]. This leads to the formation of antibiotic resistant genes (ARG) in bacteria, which can enter human body through food chain [13].

Many infections are caused by bacterial strains that are mostly commensals and sometimes opportunistic pathogens. Many antibiotic-resistant genotypes have appeared in these communal strains of microorganisms [14].

It was shown that the metabolites of acylglu-

curonide are formed by biotransformation of drugs containing carboxylic acid, are potentially chemically reactive and have hepatotoxicity, nephrotoxicity, cause hypersensitivity reactions to drugs and tissue damage by induction of immune reactions against covalent protein adducts [15].

Ciprofloxacin, like other antibiotics, when excreted from the body, can be transferred in natural environments as starting compound, hydrolysis products, conjugates and oxides [16].

Metronidazole is included in the WHO list of essential medicines as a basic drug and belongs to the group of nitroimidazoles, is effective and is used in the treatment of bacterial infections and infections caused by protozoa. [17].

Iodine is an essential trace mineral necessary for production of thyroid hormones (3,5,3',5'-tetraiodo-L-tyronine, T<sub>4</sub>; 3,5,3'-triiodo-L-tyronine, T<sub>3</sub>). Recommended daily requirement for preschool children is 90 µg, for schoolchildren – 120 µg, for adolescents and adults – 150 µg, for pregnant and lactating women – 250 µg, respectively [18]. It is known that universal salt iodization is the most effective way to eliminate cases of iodine deficiency. However, stages of its biological transformation in environmental objects have not been studied.

## Materials and methods

Studies used white outbred rats and mice of both sexes. The animals were kept in the IVC complex. Conditions of animals were in accordance with the following standards: ambient temperature was (21 ± 2) °C, humidity (50 ± 10) %, artificial light mode (12:12). All animals had free access to water and feed in *ad libitum* mode. Distribution by groups was carried out after randomization and marking of animals.

For determination of drugs biotransformation products, studies were carried out on Mature SPF-rats, which course in 7 days introduced studied drugs. Groups each with ten animals were formed: 1 group – rats receiving daily oral metronidazole in drinking water to provide a dose of 24 mg/kg per day (the recommended dose for anaerobic infections – British national form I986); 2 group – rats receiving daily intramuscularly ciprofloxacin 100 mg/kg; 3 group – oral iodine adduct in the selected dose. After 7 days, urine and feces were collected in all animals in first half of the day to determine metabolites of drugs used

in experiment. Rats were removed from the experiment in compliance with rules of laboratory animals humane treatment by decapitation after inhalation with air containing 70 % CO<sub>2</sub> at a flow rate of 30 l/min in the chamber, and biomaterial was collected.

For studying of microorganisms resistance, introduction of metronidazole, ciprofloxacin and adducts of iodine created model of sepsis in SPF mice weighing 18-20 g. Test strains *Staphylococcus aureus* ATCC 6538-P, *Escherichia coli* ATCC 8739 strain of *E. coli*; *E. faecalis* ATCC 51575; *E. faecium* ATCC 700221 at a concentration of 1.0 McF – 1 ml were chosen as infectious agents. Through 72 hours after infection was carried out blood sampling to identify priority pathogen for subsequent experiments.

Then, mice received *S. aureus* 2+ ATCC 6538-P at concentration of  $1.5 \times 10^8$  CFU/ml, in volume of 0.3 ml once intravenously, subject to aseptic rules. After 72 hours, blood was taken to re-confirm presence of pathogen. Then randomization was conducted for five animals in each group: group 1 – mice treated for 7 days with oral metronidazole in the drinking water of 24 mg/kg per day; group 2 – mice treated with daily intramuscular injection of ciprofloxacin 100 mg/kg for 7 days; group 3 – mice treated orally adduct of iodine in the selected dose and the time interval; group 4 – positive control (untreated). One week later, animals were anesthetized with subsequent cervical dislocation. For detection virulence of the strain, blood and urine were collected to determine plasma-coagulating and lecithovitellase activities, as well as to determine sensitivity to the drugs used.

Definition biotransformation substances conducted by validated methods on HPLC (Agilent 1200).

Determination of iodide ions mass concentration in 1 % aqueous solution of tetramethylammonium hydroxide was performed by inductively coupled plasma mass spectrometry (ICP-MS) [19].

Procedure for determination of antimicrobial activity was performed by method of twofold serial dilutions in physiological solution.

Preparation of suspension investigated test strains of microorganisms in physiological solution of 0.9 % NaCl by method of twofold serial dilution using inoculum of microorganism test strain at concentration of  $1.5 \times 10^6$  CFU/ml. Initial suspension of the test strain were prepared in physiological solution (0.9 % NaCl). Sterile loop

was used to select an aliquot of daily-cultured test strain, after which it was introduced into sterile tube with 5 ml of 0.9 % NaCl. Turbidity of obtained inoculum was controlled by measuring optical density on den-1 densitometer. Density of primary suspension was 0.5 Mcfarland units (0.5 McF), which corresponds to  $1.5 \times 10^8$  CFU/ml. Further, primary suspension in amount of 0.1 ml was introduced into the tube with 9.9 ml of isotonic solution to achieve working concentration of  $1.5 \times 10^6$  CFU/ml. For infection of mice, microorganisms inoculum at concentration of 1.0 units according to Mcfarland (1.0 McF) by 1 ml. Intraperitoneal method of animals infection was chosen. Animals were divided into groups according to test strains, within group according to antibiotics. Treatment was carried out in course of 7 days. For histological studies, pieces of organs measuring 10×20 mm were cut from several places (at junction of healthy and, if any, affected tissue; from Central part and from edge of organ) from the following organs: lung, liver, spleen, kidney. For doing this, excised parts of the organs were fixed in buffered 10 % solution of neutral formalin, dehydrated in ethanol solutions with an ascending concentration and poured into paraffin blocks. From paraffin blocks cut tissue into thin layers with the help of microtome was obtained sections with thickness of 5-7 μm, which were stained with hematoxylin-eosin [20, 21]. Microscopic studies of tissue sections and internal organs were performed using a direct light microscope (Leica DM1000).

Statistical processing was carried out according to standard methods [22]. Two-sided student test (t) was used to determine significance of differences between experimental and control values under normal distribution. Differences between groups were considered statistically significant at  $P \leq 0.05$ . After obtaining numerical values of the primary data, arithmetic mean (M) and standard error of mean (m) were calculated.

## RESULTS AND DISCUSSION

Results of ciprofloxacin, metronidazole and iodides concentrations determination after oral administration of medicines course (ciproxone 100 mg/kg, metronidazole 24.0 mg/kg, the adduct of iodine with 4.0 mg/kg body weight) in rats are presented in table 1.



Table 1. Concentration of drugs in urine and faeces of rats, M ± m

№	Medicinal drug	Concentration, mg/l	
		urine	faeces
1	Ciprofloxacin	157.16 ± 96.24	0.75 ± 0.35
2	Metronidazole	1086.5 ± 198.6	9.25 ± 4.5
3	Iodides	0.17 ± 0.08	9.2 ± 0.4

Determination of drugs concentration revealed their presence in biological substrates – urine and feces. Highest concentrations of antibiotics were observed in urine, and iodide ions – in feces.

Results of test strains sensitivity determination to the studied drugs under *in vitro* conditions are presented in table 2.

Table 2. Determination of test strains sensitivity to drugs *in vitro*

№	Medicinal drug, dosage	Test strains of microorganisms			
		<i>Staphylococcus aureus</i> ATCC 6538-P	<i>Enterococcus faecalis</i> ATCC 51575	<i>Enterococcus faecalis</i> ATCC 700221	<i>Escherichia coli</i> ATCC 8739
1	Metronidazol 250 mg	growth from 10 well MBC 16	growth from 2 well MBC 4000	growth from 2 well MBC 4000	growth from 11 well MBC 8
2	Ciprofloxacin (Forcip 500 mg)	growth from 12 well MBC 4	growth from 9 well MBC 32	growth from 7 well MBC 125	growth from 11 well MBC 8
3	Ciprofloxacin (Ciproxe 500 mg)	growth from 10 well MBC 4	growth from 9 well MBC 32	growth from 7 well MBC 125	growth from 11 well MBC 8
4	Iodine adduct	growth from 11 well MBC 8	growth from 11 well MBC 8	growth from 12 well MBC 4	growth from 12 well MBC 4

After intraperitoneal injection of test strains *Staphylococcus aureus* ATCC6538-P; *Escherichia coli* ATCC8739; *E.faecalis* ATCC51575; *E.feacium* ATCC700221 to mice at concentration of 1.0 McF – 1 ml infection occurred only by *Staphylococcus aureus* ATCC6538-P culture, growth of which was observed in peripheral blood. In this regard, test strain of *S.aureus* 2+ at a concentration of  $1.5 \times 10^8$  CFU/ml was selected as an infecting agent for subsequent experiments *in vivo*.

After intraperitoneal infection with *S. aureus* 2+ on the third day, course administration of drugs was started within 7 days. For isolation of microorganisms test strains we collected animal urine from the bladder to avoid contamination of test material with animals own microorganisms by sterile syringes.

For isolation of pure culture from urine, Muller-Hinton broth (MOSS) was used, followed by sowing on dense nutrient media – yolk-salt agar (JSA) after 24 hours. Grown cultures were studied for compliance with cultural, morphological, tincorial properties of *Staphylococcus aureus*. Growth of S-colonies with yellow pigment, 1-2 mm in diameter with turbidity zone (lecitovitellase activity) was observed on JSA. Microscopic preparations were prepared from the culture. Gram positive (g+) cocci, arranged in grapes were discovered. Selected culture was checked for plasmacoagulase and DNA-ase. Plasmacoagulation occurred for 4 hours. Response to DNA-ase gave positive result.

Antibiogram *S. aureus* ATCC 6538-P after treatment is presented in table 3.

Table 3. Determination of *S. aureus* ATCC 6538-P test strain sensitivity to drugs *in vivo*

№	Medicinal drug	Test strain of microorganisms <i>Staphylococcus aureus</i> ATCC 6538-P	Sensitivity alteration
1	Metronidazol	growth from 9 well (MBC 32 μ	resistance development
2	Ciprofloxacin	growth from 12 well (MBC 4 μ	no changes
3	Ciprofloxacin (Ciproxe)	growth from 12 well (MBC 4 μ	no changes
4	Iodine adduct	growth from 11 well (MBC 8 μ	no changes



In determination of sensitivity to antibiotics after treatment, it was found that there was a change in MBC to metronidazole twice towards the formation of resistance (MBC in vitro was 16  $\mu\text{g/ml}$ , in vivo was 32  $\mu\text{g/ml}$ ).

Histological findings of liver, spleen and kidney of mice infected with *S. aureus*, with application of ciprofloxacin, metronidazole and iodine adduct are shown in figures 1-10.

Mice of positive control group began to die, starting from the third day from moment of pathogen introduction. In autopsy of dying animals putrid smell was detected. Lungs were with uneven airiness, edema, and atelectasis. On lungs slice dense grainy yellowish-gray foci protruding above cut surface which when pressed secrete pus with pungent odor were observed. Liver was with not large small foci in the form of cavities with dirty yellowish color purulent contents. In heart, mainly in the ventricle myocardium was small foci with rounded shape, generally, myocardium of flabby consistency. Kidney capsule is easily removable. Kidneys were swollen of flabby consistency, on the cortical layer cut is small foci of yellowish-gray color, surrounded by red colored corolla. Spleen is cut with small yellowish-gray patches, leaving scrape on scalpel blade.

Liver of infected mice treated with ciprofloxacin consists of lymphohistiocytic infiltrates foci, mainly in portal tracts, activation of stellate reticulocytes (figure 1).

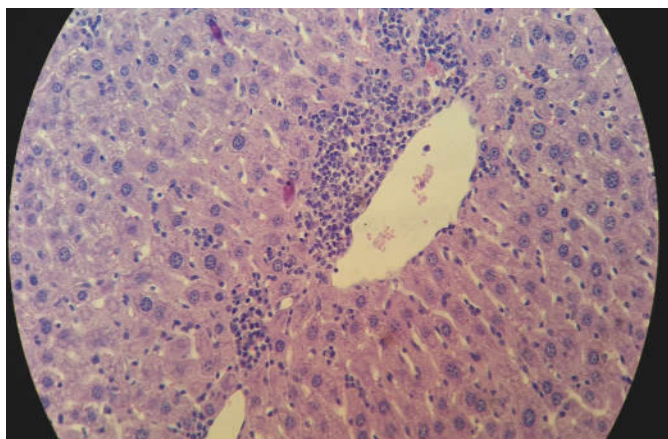


Fig. 1. Histological structure of infected mice liver, treated with ciprofloxacin. Stain: hematoxylin-eosin. Magnification:  $\times 200$

Proximal and distal tubules of kidneys are atrophied, content in lumen is mainly leukocytes. Leukocyte infiltration, spreading to the adjacent adipose tissue. Picture of purulent pyelitis (figure 2).

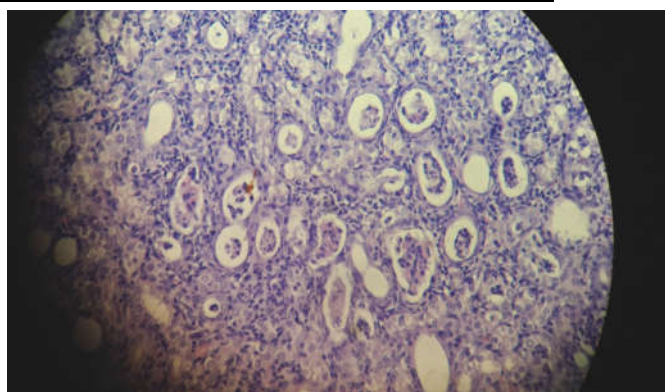


Fig. 2. Histostructure of infected mice kidneys treated with ciprofloxacin. Stain: hematoxylin-eosin. Magnification:  $\times 200$

In the spleen of infected mice treated with ciprofloxacin, giant macrophages are observed (figure 3).

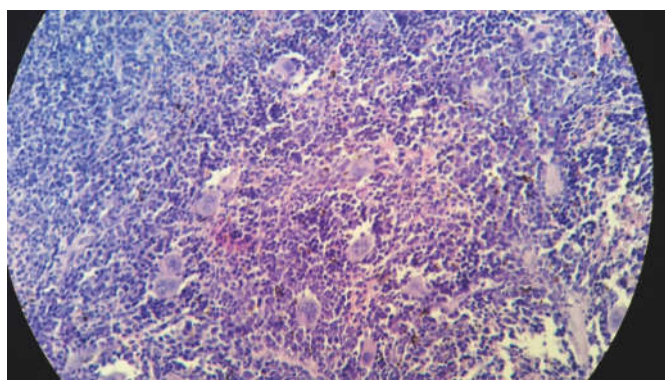


Fig. 3. Histological structure of infected mice spleen, treated with ciprofloxacin. Stain: hematoxylin-eosin. Magnification  $\times 100$

Microscopic examination of mice liver infected with *Staphylococcus aureus*, receiving metronidazole, focal purulent inflammatory infiltrates were noted in organ. Focal dystrophy of hepatocytes. Activation of Kupfer cells (figure 4).

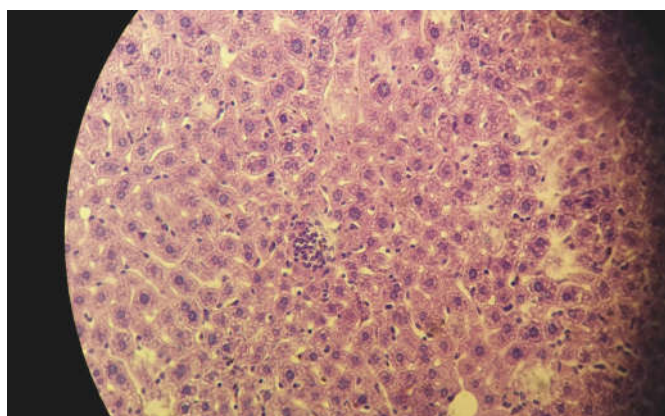


Fig. 4. Histological structure of infected mice liver receiving metronidazole. Stain: hematoxylin-eosin. Magnification  $\times 200$

In kidneys, during treatment of mice with metronidazole, focal infiltration, mainly represented by neutrophil leukocytes and colonies of bacteria is noted, infiltrates with histolysis of kidney tissue are formed. There are basophilic bacterial emboli in the vessels lumen. Picture of embolic purulent nephri-



tis (pyosepticemia). Tubules are focally atrophied, epithelium of preserved tubules is in the state of protein and large-vacuum dystrophy. In place of tubules there are unstructured areas of pink – necrotic detritus of pink color. Glomeruli are enlarged due to proliferation mesange. Also balls with epithelium proliferation of an outside leaf glomerulus capsule are noted. In individual animals is predominantly lymphomacrophagous infiltration (figures 5-6).

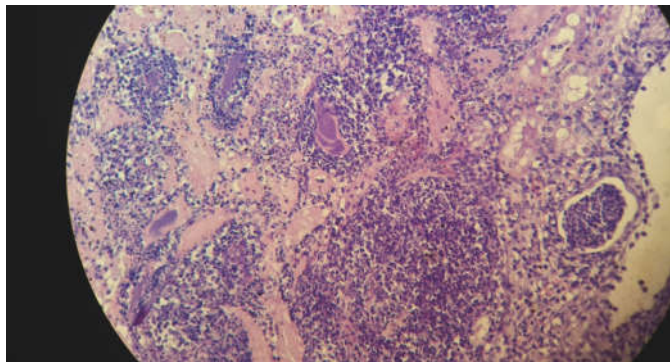


Fig. 5. Histostructure of infected mice kidneys treated with metronidazole. Stain: hematoxylin-eosin. Magnification:  $\times 200$

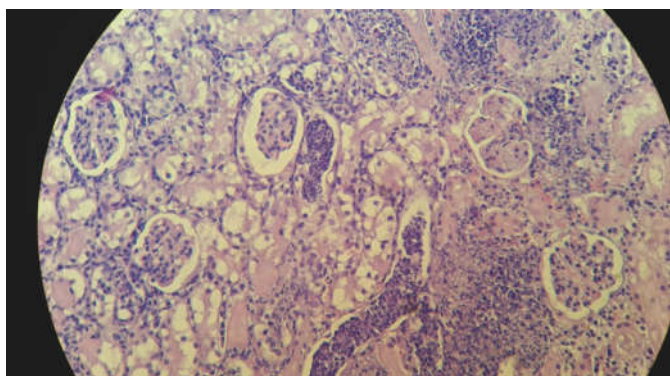


Fig. 6. Histostructure of infected mice kidneys treated with metronidazole. Stain: hematoxylin-eosin. Magnification:  $\times 200$

Spleen parenchyma pattern is erased, red and white pulp are not differentiated, lymphocytes are superseded by giant macrophages, perivascular edema and interstitial edema (figure 7).

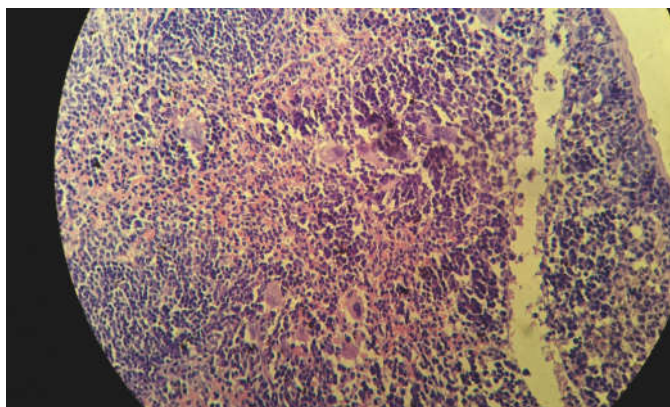


Fig. 7. Histostructure of infected mice spleen treated with metronidazol. Stain: hematoxylin-eosin. Magnification  $\times 200$

In mice treated with iodine complex, small foci of lymphohistiocytic infiltration were observed in liver. At the same time, hepatocytes are in the state of protein and small-scale dystrophy, vacuoles coalesce in individual cells. Nuclei of hepatocytes are reduced in size. Activation of stellate reticulocytes. Expansion of veins and sinusoids, hyperemia of veins (figure 8).

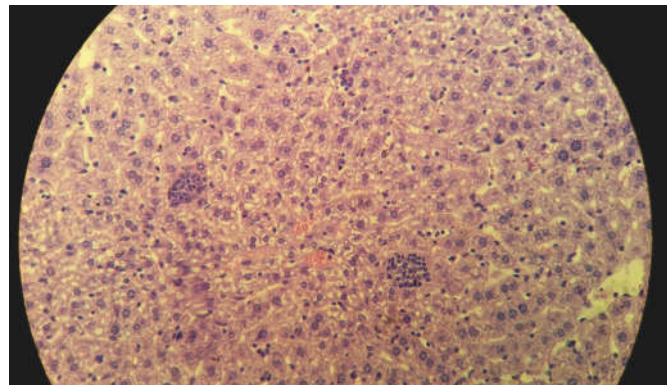


Fig. 8. Histological structure of infected mice liver treated with adducts of iodine. Stain: hematoxylin-eosin. Magnification  $\times 200$

In the kidneys there are foci of leukocyte infiltration, hemodynamic disorders. Adjacent glomeruli are enlarged due to proliferation mesange. Separate epithelial cells of distal tubules are necrobiotically changed (figure 9).

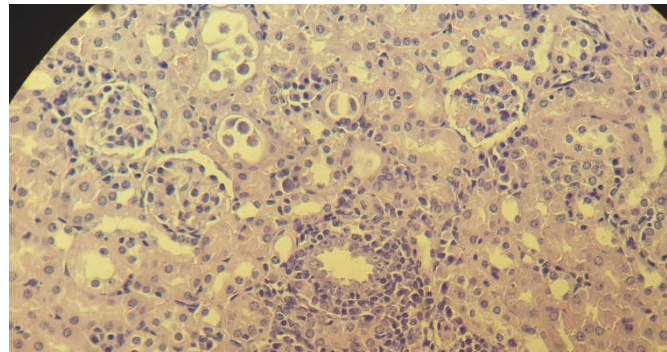


Fig. 9. Histological structure of infected mice kidneys treated with adducts of iodine. Stain: hematoxylin-eosin. Magnification  $\times 200$

Giant macrophages are observed in the spleen (figure 10).

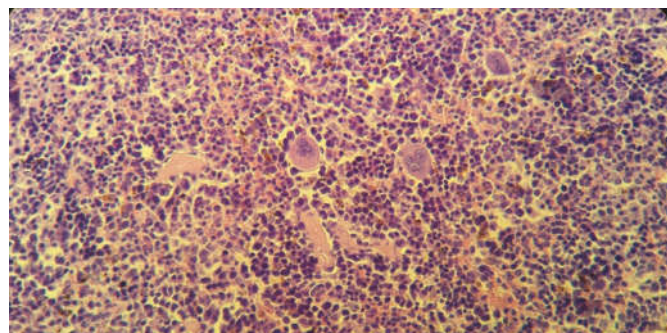


Fig. 10. Histological structure of infected mice spleen treated with adducts of iodine. Stain: hematoxylin-eosin. Magnification  $\times 200$

## CONCLUSION

Validation of sample preparation method and methods of ciprofloxacin and metronidazole analysis were carried out. According to parameters of accuracy, precision, repeatability, intermediate precision, LOD, LOQ, linearity and operating range, procedure is validated, and results of validation fully meet validation plan requirements. Injection of drugs to rats (ciprofloxacin, metronidazole, iodine), which are most widely used in medical practice, with course application for 7 days, leads to detection of biotransformation products in urine and faeces. Concentration of ciprofloxacin and metronidazole in urine was significantly higher than in feces, in contrast, concentration of iodides in feces is greater than in urine.

In determination of *Staphylococcus aureus* ATCC 6538-P, *Enterococcus faecalis* ATCC 51575, *Enterococcus faecalis* ATCC 700221, *E. coli* ATCC 8739 strains *in vitro* sensitivity, they all showed sensitivity to the tested drugs. With intraperitoneal administration of these strains to mice, infection occurred only *S. aureus* ATCC 6538-P at concentration of  $1.5 \times 10^8$  CFU/ml, which was determined both in blood and urine. *S. aureus* ATCC 6538-P caused in animals by histological research of septicopyemia picture. Use of metronidazole, ciprofloxacin and iodine adducts led to positive dynamics of infectious process, but pathogen was found in urine after therapy end. Treatment of *S. aureus* ATCC 6538-P mice infected with studied drugs led to formation of resistance to metronidazole (MBC *in vivo* is twice as much as MBC *in vitro*, 32 µg/ml and 16 µg/ml, respectively). This fact indicates that infectious agent can enter wastewater and environment in resistant form.

Thus, studies showed the need for further study of drugs biotransformation products receipt problem in the environment of urban areas, followed by the risk of conditionally pathogenic microorganisms resistance.

We believe that the principle of taking into account total residue of drugs in wastewater in the assessment of risks to the environment is not sufficient [23], since it assumes that the drugs themselves exhibit same effects in the environment. However, transformed drugs may exhibit whole range of new toxicological effects than the original compound. Many pharmaceutical products and their metabolites can be removed only partially or not at all in

the process of traditional wastewater treatment technologies, which leads to their accumulation in soils and agricultural products, further along food chain can enter human body and as a result form microorganisms resistance to the used drugs. Since drug resistance is currently a global health problem, there is the need to include biotransformed pharmaceuticals (metabolites) in open water sources monitoring programs of the Republic of Kazakhstan.

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## ЗЕРТХАНАЛЫҚ ЖАНУАРЛАРДА АНТИБАКТЕРИАЛДЫҚ ПРЕПАРАТТАРДЫҢ ТРАНСФОРМАЦИЯЛАУ ӨНІМДЕРІН ЖӘНЕ МИКРОБИОЛОГИЯЛЫҚ БЕЛСЕНДІЛІГІН ЗЕРТТЕУ

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### ТҮЙІН

Бастапқы қалпына қарағанда, трансформацияға ұшыраған дәрі-дәрмектер кешенді жаңа токсикологиялық әсерлер көрсетуі ықтимал. Пайдаланылған суларды дәстүрлі тазалау барысында көптеген фармацевтикалық құралдар мен олардың метаболиттері тек жартылай немесе мүлдем тазаланбайтындықтан олар топырақта және ауыл шаруашылығы өнімдерінде жинақталады. Онан әрі тізбектеліп барып адам организміне өтетіндіктен, пайдаланылып жүрген дәрілік препараттарға төзімділіктерді қалыптастырады. Тәжірибе барысында ципрофлоксацин, метронидазол және аддукт иодының зертханалық жануарлар (егеуқұйрық) ағзаларына әсеріне зерттеулер жүргізілді. *Staphylococcus aureus*, *Enterococcus faecalis* және *Escherichia coli* микроағзалар штамдарының *in vitro* жағдайында пайдаланылып жүрген препараттарға сезімділіктері анықталды. Тышқандарға сепсис жасау арқылы микроағзаның сезімталдығының өзгеруін зерттедік. Ципрофлоксацин, метронидазол мен иод аддуктінің биотрансформацияға ұшыраған өнімдері жүйелі қолданғанда атжалмандардың несептері мен үлкен дәретінде 7 күннен соң көріне бастағанын байқадық. Ципрофлоксацин мен метронидозолдың аздаған мөлшерінің жиынтығы несептерінде, алиодидтің аздаған мөлшерінің жиынтығы үлкен дәретінде анықталды. *In vitro* зерттеулер барысында *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli* микроағзалардың штамдары зерттеліп отырған препараттарға сезімталдық байқатты. Тышқандарды метронидазол, ципрофлоксацин және иод аддуктімен емдеу септикопиемияның ағымды динамикасында жақсы нәтиже бергенімен, метронидазолға тұрақтылықты қалыптастыруға алып келді. Зерттеу нәтижесі биотрансформацияға ұшыраған фармацевтикалық препараттарды (метаболиттерді) Қазақстан Республикасының ашық су көздеріне мониторинг жасау бағдарламасына енгізудің қажет екендігін көрсетті.

Негізгі сөздер: фармацевтикалық препарат, ципрофлоксацин, метронидазол, иод аддукті, биотрансформация, қоршаған орта, резистенттілік, микроағзалардың тест-штаммы, зертханалық жануар, сепсис, морфоструктура.



## ИЗУЧЕНИЕ ПРОДУКТОВ ТРАНСФОРМАЦИИ И МИКРОБИОЛОГИЧЕСКОЙ АКТИВНОСТИ АНТИБАКТЕРИАЛЬНЫХ ПРЕПАРАТОВ У ЛАБОРАТОРНЫХ ЖИВОТНЫХ

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### АБСТРАКТ

Трансформированные лекарственные средства могут проявлять целый комплекс новых токсикологических эффектов, чем исходные соединения. Многие фармацевтические средства и их метаболиты удаляются только частично или вообще не удаляются в процессе традиционных технологий очистки сточных вод, что приводит к их накоплению в почвах и сельскохозяйственной продукции. Далее по пищевой цепи они могут поступать в организм человека и в результате формировать устойчивость микроорганизмов к применяемым лекарственным препаратам. Изучали биотрансформация лекарственных веществ: ципрофлоксацина, метронидазола и аддукта иода в организме лабораторных животных (крыс). Определяли чувствительность штаммов микроорганизмов *Staphylococcus aureus*, *Enterococcus faecalis* и *Escherichia coli* к используемым препаратам в условиях *in vitro*. Изучали изменение чувствительности микроорганизмов при моделировании сепсиса у мышей. Показано, что продукты биотрансформации ципрофлоксацина, метронидазола и аддукта иода обнаруживаются в моче и фекалиях крыс при их курсовом применении в течение 7 дней. Наибольшая концентрация ципрофлоксацина и метронидазола обнаружена в моче, а иодидов – в фекалиях. В исследованиях *in vitro* показано, что штаммы микроорганизмов *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli* проявляли чувствительность к исследуемым препаратам. Лечение мышей метронидазолом, ципрофлоксацином и аддуктом иода вызывало положительную динамику течения септикопиемии, однако приводило к формированию устойчивости к метронидазолу. Проведенные исследования показали необходимость включения биотрансформированных фармацевтических препаратов (метаболитов) в программы мониторинга открытых водоемов Республики Казахстан.

Ключевые слова: фармацевтический препарат, ципрофлоксацин, метронидазол, аддукт иод, биотрансформация, окружающая среда, резистентность, тест-штамм микроорганизма, лабораторное животное, сепсис, морфоструктура.