MONITORING OF SALMONELLA INFECTION OF POULTRY FOR THE PERIOD FROM 2016 TO 2020

Zhanna Klishchova
Department of Veterinary Examination, Microbiology, Zoohygiene and Safety and Quality of Livestock Products
kgejp1990@gmail.com

Svetlana Nazarenko
Department of Veterinary Examination, Microbiology, Zoohygiene and Safety and Quality of Livestock Products
nazarenko sveta2014@gmail.com

1Sumy National Agrarian University
160 Herasyma Kondratieva str., Sumy, Ukraine, 40021

Abstract
Salmonellosis is an acute intestinal infectious disease that belongs to the group of zoonoses that cause acute toxic infections in humans through the consumption of products of animal or plant origin contaminated with bacteria of the genus Salmonella spp. Salmonellosis is registered in all countries of the world, and our state is no exception.

The aim of the research. Monitor salmonellosis pathogens in Ukrainian poultry farms and, on the basis of the obtained data, establish the relationship between avian and human diseases for salmonellosis.

Materials and methods. The research was conducted in poultry farms in the North-Eastern region of Ukraine. Test systems from R-biopharm-Compact Dry SL and RIDA® STAMP SL were used for rapid diagnosis of bacterial microflora. To establish the salmonella serotype, the Spectate® salmonella test system was used, which is based on the use of latex strips coated with special antibodies to the corresponding serogroups of Salmonella A, B, C, D. Microbiological examination was performed washing from the walls, floor, eggshell, Brooder’s cabinets and pathogens heart, liver, gallbladder of the dead bird. The relationship between isolated salmonella serovars from poultry was compared with salmonella isolated from humans according to the reporting form No. 40 «Zdorov».

Results. Various salmonella serovars were isolated from experimental farms of Ukraine, namely:
- Serovars such as S. Gallinarum-pullorum – 7.9 %, S. Enteritidis – 6.5 % were isolated from the meat and egg direction. From the meat direction, the following are: S. Tiphimurium – 12.5 %, S. Arizona – 6.0 %, S. Enteritidis – 3.5 %. From the breeding direction were also isolated from adult birds and hatching eggs in large quantities – S. Gallinarum-pullorum – 19.0 %, S. Tiphimurium – 17.8 %, S. Enteritidis – 10.1 %.
- The following species were separately isolated from the premises of the same farms in percentage terms: S. London – 1.7 %, S. Infantis – 1.5 %, S. Bredeney – 1.4 %, S. Teiogues – 1.4 %, S. Jawa – 1.2 %, S. Montevideo – 1.1 %, and 1 % each isolated S. Kentiki, S. Abony and S. Oxford. Pathogens of paratyphoid diseases were isolated from poultry and poultry products (S. Gallinarum-pullorum, S. Enteritidis, S. Tiphimurium) for the entire study period from 2016 to 2020 inclusive.
- Comparing the obtained data from state institutions of humane medicine, it should be noted that there was a tendency for the spread of salmonellosis among people associated with the consumption of poultry products.

Conclusions. The obtained results indicate that salmonella infection is quite common among a number of poultry farms of different technological direction. In particular, bacteria of the genus Salmonella were isolated from meat, eggs and birds of different ages from the studied poultry houses in the North-Eastern region of Ukraine.

As a result of monitoring of salmonellosis in Ukraine over the past five years, fluctuations in the incidence of salmonellosis among people ranged from a maximum of 74 % (2018) to a minimum of 30.1 % (2020), and for two years, there has been a reduction in cases due to the introduction of new international requirements and standards for product quality control at all stages of production of the Hazard Analysis and Critical Control Point (HACCP) system.

Keywords: Salmonella, salmonellosis, food poisoning, rare, serotypes, poultry, humans, poultry farm, food.

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1. Introduction
The term «salmonellosis» currently includes a large group of clinically polymorphic diseases caused by various serotypes of bacteria of the genus Salmonella of the family Enterobactereaceae [1]. The causative agents of salmonellosis mainly affect all species of animals. But the severity of the
disease depends on the serotype, susceptibility of animals, the form of the course, and a number of other factors that contribute to the spread of this problem [2].

Outbreaks appear to be exacerbated throughout the world through the consumption of animal products. The Centers for Disease Control and Prevention estimates that about 1.35 million cases of salmonellosis are reported each year in the United States. Among them, 26,500 people [3] were hospitalized with pronounced clinical signs and 420 deaths were registered. The countries of Africa and Southeast Asia are no exception, the incidence is 129 people per 100 thousand population. The estimated incidence was 1334 cases per 100 thousand population in Libya [4] mainly affected the age group of people 20–39 years without significant gender differences [4].

According to the Public Health Center of the Ministry of Health of Ukraine, as of August 27, 2020, 16 outbreaks of salmonellosis have been registered since the beginning of the year (Zaporizhia region – 4, Ivano-Frankivsk region – 2, Vinnytsia, Dnipropetrovsk, Zakarpattia, Kirovohrad, Odesa, Poltava, Rivne, Sumy, Cherkasy region and Kyiv – 1), as a result of which 130 people were injured (including 29 children under 15) [5]. The issue of elimination of salmonellosis among humans and animals remains open due to the significant circulation of salmonella in the environment [6] and the rapid acquisition of antibiotic resistance to most drugs used [7, 8].

Therefore, the aim of the research was to determine the causative agents of salmonellosis in poultry farms of Sumy region of different technological direction. And to analyze statistics in general in Ukraine and Sumy region on the incidence of salmonellosis in humans and poultry.

2. Methods and materials

The research was conducted in poultry farms of Ukraine. Test systems from R-bio- pharm-Compact Dry SL and RIDA®STAMP SL were used for rapid diagnosis of bacterial microflora. To establish the salmonella serotype, the Spectate® salmonella test system was used, which is based on the use of latex strips coated with special antibodies to the corresponding serogroups of Salmonella A, B, C, D. Microbiological examination was performed on washings from the walls, floor, eggshell, Bruder’s cabinets and pathogens heart, liver, gallbladder of the dead bird. Selected pieces of parenchymal organs under sterile conditions were ground in mortars with a small amount of saline, followed by seeding in one of the enrichment media (magnesium or selenite) in a ratio of 1:5, with simultaneous reseeding on the differential diagnostic medium. Each experimental fraction of bile was sown in vials with slightly alkaline nutrient broth in a ratio of 1:10 and on a differential nutrient medium. In 18–24 hours from the vials were made re-seeding on differential diagnostic media. In the case of negative results, sowing was repeated for 3, 5, 7 days, using weakly selective media. Studies of biological material were carried out taking into account the requirements of DSTU ISO 18593:2006 «Microbiology of food and animal feed».

The analysis for salmonellosis was established on the basis of the analysis of epizootological inspection of poultry farms and adjacent areas, clinical signs, results of pathological autopsy and results of serological, bacteriological researches, pathological material from birds of different age groups [9]. The relationship of isolated salmonella serovars from poultry was compared with salmonella isolated from humans according to the reporting form No. 40 «Zdorov». Statistical analysis of the results was performed using the program for analysis and visualization of data STATISTICA-7 and Excel 2010 (Microsoft Office 2010). The presence of differences between the studied indicators was assessed using the Student’s test.

3. Results

Various salmonella serovars were isolated from experimental poultry farms of the North-Eastern region of Ukraine, namely:

Serovars such as S. Gallinarum-pullorum – 7.9 %, S. Enteritidis – 6.5 % were isolated from the meat-egg direction.

From the meat segment there were isolated: S. Tiphimurium – 12.5 %, S. Arizona – 6.0 %, S. Enteritidis – 3.5 %.

S. Gallinarum-pullorum – 19.0 %, S. Tiphimurium – 17.8 %, S. Enteritidis – 10.1 % were also isolated from the breeding direction from adult birds and hatching eggs.
The following species were separately isolated from the premises of the same farms in percentage terms: *S. London* – 1.7 %, *S. Infantis* – 1.5 %, *S. Bredeney* – 1.4 %, *S. Tsioque* – 1.4 %, *S. Jawa* – 1.2 %, *S. Montevideo* – 1.1 %, and 1 % of each *S. Kentyki*, *S. Abony* and *S. Oxford* was isolated. Pathogens of paratyphoid diseases from poultry and poultry products (*S. Gallinarum-pullorum, S. Enteritidis, S. Tiphimurium*) were isolated for the entire study period from 2016 to 2020.

Comparing the obtained data from state institutions of humane medicine, there was a tendency to the spread of salmonellosis (Table 1).

**Table 1**

Survey of people for infectious diseases for the period 2016–2020

<table>
<thead>
<tr>
<th>Infectious diseases</th>
<th>Year of registration</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>2016</td>
</tr>
<tr>
<td>Typhoid fever and paratyphoid fever</td>
<td>21118</td>
</tr>
<tr>
<td>Salmonella infections</td>
<td>2863</td>
</tr>
<tr>
<td>– y %*:</td>
<td>33</td>
</tr>
<tr>
<td>Food poisoning is caused by shigellosis and yersiniosis</td>
<td>1115</td>
</tr>
<tr>
<td>– y %*:</td>
<td>16.9</td>
</tr>
<tr>
<td>Other acute intestinal infections</td>
<td>5829</td>
</tr>
<tr>
<td>– y %*:</td>
<td>53.2</td>
</tr>
</tbody>
</table>

Note: * – the following formula was used to calculate the total incidence rate \( m\% = \left(\frac{n_1 \times 1000}{n_2}\right) \times 100 \), where \( m\% = \frac{17826\times1000}{2637} = 67.59 \), \( n_1 \) – number of patients and suspected diseases, \( 1000 \) – indicator per 1000 population, \( n_2 \) – number of patients with laboratory-confirmed diagnosis.

Salmonellosis is a significant percentage of the total number of officially laboratory-confirmed diagnoses of intestinal diseases registered in Ukraine. Over a period of 4 years, fluctuations in the incidence of salmonellosis were observed. Since 2016, the number of patients and suspects of the disease was 2,863 people, of whom 854 (33 %) were officially ill. In 2017, 2,637 cases were registered (67 %), in 2018 – 2006 (75 %), in 2019, 250 cases were detected, which is 41 % of the total number of suspects in the disease. Compared to 2020, the percentage of salmonellosis was lower than last year and was only 30 %. That is, in the last two years (2019 and 2020) there was a tendency to reduce the incidence rate compared to the period 2016-2018 (from 33 % to 74 %). Regarding food poisoning caused by shigellosis and yersiniosis in 2017, the percentage is 24.1 % (979 officially registered cases of human disease per 1000), in 2018 – 32 % (835 per 1000) and 726 (30 %) cases have been registered in the last two years. This problem remains relevant not only among humans, but also among birds, as the prevention of salmonellosis should be comprehensive and include a wide range of veterinary, sanitary and anti-epidemic measures. But, poultry owners ignore such simple rules, as evidenced by the monitoring. Serovars *S. Kentyki*, *S. Abony* and *S. Oxford* are not typical for this area of the North-Eastern region of Ukraine. The first data on their detection were registered in 2016, but only from the premises of birds. Diseases in chickens and poisonings in humans have not been reported so far. It should be noted that toxicoinfections in humans were caused during the consumption of various contaminated products. The data are shown in Fig. 1.

According to the diagram (Fig. 1), most often food poisoning in humans was caused by the consumption of poultry products, they account for 68.1 %. These included isolated and exotic Salmonella (*S. Enteritidis, S. Tiphimurium, S. Kentyki, S. Abony, and S. Oxford*). In second place are poisonings caused by the consumption of confectionery, their number is 19.7 %. This is due to the use of the main ingredient of egg melange, which is used to produce various fillers, but in violation of the technical conditions of production of bakery products, all products are contaminated by foreign microflora.
Fig. 1. Percentage distribution of isolated salmonella that caused food poisoning in humans

4. Discussion
Various livestock enterprises, poultry houses and their objects are always subject to monitoring research [9]. As a result of these researches various microflora which is conditionally pathogenic or on the contrary pathogenic is always isolated. Representatives of opportunistic pathogens are present in each species of microorganisms [10, 11]. The family Enterobacteriaceae is no exception and includes more than 20 genera that cause various infectious diseases not only in birds, but also in humans [12, 13]. Investigating poultry farms of different technological directions, most isolates of Salmonella Enterica serovar Gallinarum biovar Gallinarum [13] were isolated from poultry and poultry products. The main cause of outbreaks of typhoid infection in birds is difficult to establish as it can be brought in from outside by service personnel. Outbreaks of typhoid infection have not been officially registered due to the limitations of modern diagnostic methods or due to the neglect of the rules of preventive measures by farms [14, 15]. However, salmonellosis remains a serious economic problem for livestock in countries where control measures are not effective, or in countries where climatic conditions contribute to the spread of these microorganisms in the environment [16]. Over the past 5 years, there has been an explosion of genetic and immunological information about the biology of these pathogens, which have adapted well to the conditions of the environment and the body of birds and humans. This is evidenced by the annual epidemiological outbreaks of salmonellosis among people registered in Ukraine in large cities [17]. Analyzing the data for the last 5 years, salmonellosis were registered among people with different serotypes of exotic (Enteritidis, S. Tiphimurium, S. Kentyki, S. Abony and S. Oxford) salmonella, which were contained in meat and poultry products. This factor of transmission through consumer products puts them at the level of socio-economic problems that require constant monitoring by the relevant services [18, 19].

Study limitations. The limitation of the research is the small number of surveyed poultry and limited access to the statistics of the Ministry of Health on cities, districts, regions and Ukraine as a whole.

Prospects for further research. To carry out constant monitoring of the circulating microflora of Ukrainian poultry farms for avian salmonellosis. To establish the species spectrum of isolated microorganisms from the air, technological equipment of poultry houses and hatcheries, etc. To determine the sensitivity of Salmonella serotypes to antibacterial drugs, disinfectants, that officially registered in Ukraine.

5. Conclusions
1. The obtained results indicate that salmonella infection is quite common among a number of poultry farms of different technological direction. In particular, from the studied poultry farms of the North-Eastern region of Ukraine, bacteria of the genus Salmonella were isolated from meat, eggs and birds of different ages.

2. As a result of monitoring of salmonellosis in Ukraine over the past five years, fluctuations in the incidence of salmonellosis among people ranged from a maximum of 74 % (2018) to a minimum of 30.1 % (2020), in the last two years there has been a decrease in cases due to the introduction of new international requirements and standards for product quality control at all stages of production of the HACCP system.

Conflicts of interest
The authors declare that they have no conflicts of interest.
References


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