



Spatio-temporal analysis of leptospirosis in cattle in Ukraine over 2005–2024

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Leptospirosis is a zoonotic infection caused by genetically diverse and continuously evolving pathogens, posing a significant threat to both animal and human health. This study investigates the prevalence of *Leptospira* spp. in cattle populations in Ukraine over the period 2005–2024, encompassing both clinically apparent and subclinical infections. During this period, a total of 2 924 864 cattle serum samples were tested by veterinary laboratories in Ukraine for leptospirosis and 148 850 were positive, which accounted for 5.1%. In general, the number of tested samples decreased significantly (the number of tested samples in 2024 was only 12.2% of the initial one – reduction of studies by 8.18 times from 2005). The seroprevalence was highest in 2005 and 2007 – 8.6% and 7.2%, respectively. The minimum level of seroprevalence was observed in 2022 and 2024 – 1.8% and 1.6%, respectively. During 2009–2018, the incidence was at approximately the same level (seroprevalence in the range of 3.6–4.7%), and later this figure began to decline again – from 4.2% in 2018 to 1.6% in 2024. During the research period, there was a steady downward trend in the number of seropositivity for bovine leptospirosis. Studies have shown that there was a decrease in the number of positive reactions to *Leptospira* serovars *kabura*, *polonica* and *tarassovi*, but an increase in the number of positive reactions to *copenhageni*, *canicola* and *bratislava* serovars. The percentage of seroprevalence of *Leptospira grippityphosa* and *pomona* serovars in cattle remained at the same levels. It is established, that the very high-risk area for bovine leptospirosis in Ukraine includes five regions: Donetsk, Cherkasy, Kharkiv, Sumy, and Dnipropetrovsk. The total number of positive samples in this zone is 91 318, which is 61.3%. The least risky regions in terms of bovine leptospirosis are Ivano-Frankivsk, Luhansk, Crimea, Zakarpattia and Lviv regions (only 1.3% or 1 475 positive samples). Cartographic analysis of the circulation of diagnostic leptospira serovars among cattle in Ukraine indicates the heterogeneity of the epizootic situation regarding this disease in different regions of our country. Our studies have shown that in the etiological structure of bovine leptospirosis in Ukraine the dominant serovars are *kabura* (serogroup *Hebdomadis*) (12.2%), *polonica* (serogroup *Sejroe*) (9.9%), *copenhageni* (serogroup *Icterohaemorrhagiae*) (5.7%), *tarassovi* (serogroup *Tarassovi*) (4.4%). Positive reactions to other *Leptospira* serovars were rarely observed: *bratislava* (serogroup *Australis*) (2.8%), *grippityphosa* (serogroup *Grippityphosa*) (2.2%), *pomona* (serogroup *Pomona*) (1.3%) and *canicola* (serogroup *Canicola*) (1.3%).

Keywords: zoonotic disease risks; dynamics of livestock leptospirosis; serovar prevalence shifts; geospatial analysis; pathogen sources.

Introduction

Leptospirosis is the most widespread zoonosis in the world; it is present on all continents except Antarctica, and evidence of *Leptospira*'s carriage has been found in almost all mammalian species studied (Garcia-Lopez et al., 2023). A significant number of outbreaks of this disease are reported worldwide after natural disasters, mainly in urban areas of Guyana (Dechet et al., 2005), India, Italy, Indonesia, Malaysia and the Philippines (Amilasan et al., 2012). A significant number of outbreaks of endemic leptospirosis are reported in Thailand (Chadsuthi et al., 2021), Italy, New Caledonia (Goarant et al., 2009), Vietnam, the Netherlands, Brazil (Galan et al., 2021), Laos, Mexico, Germany (Schmidt et al., 2021), New Zealand, Argentina, Sri Lanka (Warnasekara et al., 2019), California (Meites et al., 2004), Nicaragua, India (Antima & Banerjee, 2023). Leptospirosis has been a global problem for many decades.

Currently, phylogenetic classification differentiates *Leptospira* species based on DNA relatedness (Vincent et al., 2019), and 68 genetic species are currently recognized (Sohm et al., 2023). This classification coexists with a historical, serological classification that recognizes more than 300 *Leptospira* serovars grouped into serogroups (Picardeau, 2017) based on lipopolysaccharide (LPS) expression.

The disease affects the reproductive function of animals and causes significant economic losses. The disease is caused by pathogenic bacteria of the genus *Leptospira* (Desa et al., 2021). Wild and domestic animals are carriers of these pathogenic microorganisms (Fischer et al., 2018; Harran et al., 2023). In most mammals, leptospires persist in the convoluted renal tubules and are excreted in the urine. Several mammalian species, mainly rodents, are the main reservoirs of the pathogen, and dogs, cattle, and pigs are frequent carriers. Dairy cattle also harbor the pathogen (Desvars et al., 2013; Win et al., 2024). Vertical transmission from infected cattle to the fetus contributes to the spread of the pathogen (Otaka et al., 2013; Nogueira et al., 2020). Humans often become infected after contact with soil or water contaminated with the pathogen from the urine of carrier animals (Harran et al., 2024). Water sports and occupations involving contact with animal carriers are risk factors, and outbreaks are often unpredictable (Munoz-Zanzi et al., 2020). Mammals, including cattle, are occasional or permanent hosts of various pathogenic serologic variants (Ellis, 2015). Leptospires are frequently in a commensal relationship with the host, causing little or no harm (Bierque et al., 2020; Antima & Banerjee, 2023). Thus, a set of reservoir animals contributes to environmental contamination and ensures relay transmission of the pathogen to animals and humans (Viana et al., 2014).

The disease is considered systemic in terms of organ damage and often leads not only to significant economic losses, but also to the death of animals and humans (Harran et al., 2024). The genus *Leptospira* is currently divided into 69 genomic species, including saprophytic and pathogenic bacteria (Vincent et al., 2019; Fernandes et al., 2022). Eight pathogenic species are predominantly involved in infecting humans and animals (Vincent et al., 2019), which in turn are divided into 26 serogroups (Harran et al., 2024). Each year, mainly *L. kirschneri* and *L. interrogans* cause approximately 1 million human cases and 60,000 deaths worldwide (Costa et al., 2015; Fiecek et al., 2017; Picardeau, 2017; Guillois et al., 2018), with most deaths occurring in countries with extremely limited resource capacity (Pakoa et al., 2018). Consequently, 1.03 million people are infected annually (95% confidence interval (CI) 0.43 to 1.75 million) and 58 900 people die (95% CI 23 800 to 95 900). Of these, a significant proportion of those infected (48%, 95% CI 40 to 61) and deaths (42%, 95% CI 34 to 53) are adults aged 20 to 49 years (Costa et al., 2015). Leptospirosis carries risks and economic burden that are fairly compared to the leishmaniasis, schistosomiasis, or lymphatic filariasis (Torgerson et al., 2015). According to the US Centers for Disease Control and Prevention (CDC), the mortality rate is between 5% and 15% among people with severe disease and more than 50% among people with severe pulmonary hemorrhage (Win et al., 2024). And such a significant annual mortality rate from leptospirosis is five times higher than the number of deaths from Ebola, which was recorded in 2013–2016 (Shultz et al., 2016).

Leptospirosis in cattle is mainly caused by serovar *hardjo*, in which two species are identified, *L. interrogans* serovar *hardjo* (type *Hardjoprajitno*) and *L. borgpetersenii* serovar *hardjo* (type *Hardjobovis*), the latter being the most common worldwide (Loureiro & Lilenbaum, 2020). The two species are antigenically similar because the RFB locus, which contains genes encoding proteins involved in LPS biosynthesis (De La Peña-Moctezuma et al., 2001). There is a study of a deterministic mathematical model that describes the dynamics of leptospirosis transmission in a cattle herd, including asymptomatic and vaccinated animals, which is an absolute burden for farmers and farms, moreover, it is a constant zoonotic hazard (Regassa & Obsu, 2024). Clinical signs of leptospirosis in cattle are mostly characterized by reproductive problems (infertility, abortion, sudden decrease in milk production and underdevelopment of offspring). In infected calves, an acute, more severe form of the disease is observed, which can cause death (Gelalcha et al., 2021).

Leptospire transmission occurs primarily through direct or indirect (i.e., through contaminated water or soil) contact with the urine of infected animals (Munoz-Zanzi et al., 2020), although sexual transmission is also possible (Loureiro & Lilenbaum, 2020). The bacteria can enter the body of a susceptible animal through mucous membranes or damaged skin. After the leptospiraemic phase, the bacteria can colonize various organs, especially the kidneys, from where they then periodically appear in the urine (Sohm et al., 2023) or genital tract, so they are occasionally found in semen (Loureiro & Lilenbaum, 2020).

Often, the vast majority of infected cattle remain asymptomatic carriers and shed the pathogen in the urine for a long time (sometimes up to 3 years), which primarily depends on the serovar (Roqueplo et al., 2013; Rajeev et al., 2014; Ibrahim et al., 2022; van den Brink et al., 2023; Monti et al., 2023). Colonization of the reproductive system by the pathogens often leads to renal failure and reproductive problems in animals carrying the pathogen (Regassa & Obsu, 2024). The transmission of the leptospirosis pathogens in cattle herds is prevented by vaccination, antibiotic treatment of animals, and disinfection. Vaccination in the system of control measures for bovine leptospirosis ultimately prevents the risk of human infection with leptospire (Sanhueza et al., 2018). It has been estimated that vaccination against this disease provides cattle with active protection within 84% and prevents asymptomatic leptospirosis by about 88% (Bergmann et al., 2022).

We have analyzed the serological prevalence of leptospirosis among cattle in Ukraine for the period 2005–2024, and identified the most epizootologically significant serovars of leptospire.

Materials and methods

The current study presents a retrospective epidemiological assessment of leptospirosis incidence and the etiological structure of infections in cattle across different regions of Ukraine between 2005 and 2024. Data were sourced from official reports provided by regional laboratories operating under the State Service of Ukraine on Food Safety and Consumer Protection (SSUFSCP), as well as from research conducted by the State Research Institute of Laboratory Diagnostics and Veterinary and Sanitary Expertise (SRILDVSE) in Kyiv, Ukraine. Information extracted from the 1–Vet and 2–Vet reporting forms for the specified years was compiled and subjected to systematic analysis. Seroprevalence rates for each oblast were determined by calculating the ratio of leptospirosis-positive samples to the total number of samples tested in that region. All cases included in the analysis were confirmed through the laboratory diagnostics, specifically via the microscopic agglutination test (MAT), conducted in accredited institutions.

The cultures of the reference *Leptospira* strains including eight serovars, were used to perform the MAT (Table 1). These diagnostic strains of *Leptospira*, used in serological studies of leptospirosis in animals, were cultivated and prepared by the veterinary diagnostic laboratories in Ukraine. The leptospire were cultivated in a Korthof liquid medium at 28–30 °C under aerobic conditions.

Table 1

List of the diagnostic strains of leptospire

Number	Serogroup	Serovar	Strain
1	<i>Hebdomadis</i>	<i>kabura</i>	<i>Kabura</i>
2	<i>Sejroe</i>	<i>polonica</i>	<i>493 Poland</i>
3	<i>Icterohaemorrhagiae</i>	<i>copenhageni</i>	<i>M 20</i>
4	<i>Tarassovi</i>	<i>tarassovi</i>	<i>Perpelicyni</i>
5	<i>Australis</i>	<i>bratislava</i>	<i>Yež bratislava</i>
6	<i>Grippotyphosa</i>	<i>grippotyphosa</i>	<i>Moskva V</i>
7	<i>Pomona</i>	<i>pomona</i>	<i>Pomona</i>
8	<i>Canicola</i>	<i>canicola</i>	<i>Hond Utrecht IV</i>

The diagnostic procedure followed the guidelines set out in the World Organisation for Animal Health (WOAH). In brief, serum samples were initially diluted at a 1:25 ratio and then mixed in equal volumes with live cultures of various *Leptospira* serovars, resulting in a working dilution of 1:50 for the preliminary screening. Samples exhibiting agglutination with one or more serovars during this initial stage were subjected to serial twofold dilutions to determine the end-point titre corresponding to 50% agglutination (Petrakovsky, 2021). According to the criteria for unvaccinated animals, samples demonstrating titres of 1:50 or higher were interpreted as positive. In compliance with national veterinary standards (specifically, the Instructions on preventive measures and treatment of animals for leptospirosis) serological testing was performed using MAT at the following dilution levels: 1:50, 1:100, 1:500, and 1:2500. Titres exceeding 1:2500 were not assessed. All serum samples included in the study originated from cattle that had not been vaccinated against leptospirosis.

In instances where a sample showed positive reactions to multiple serovars, interpretation followed Ukrainian diagnostic protocols: the serovar associated with the highest antibody titre was considered the causative agent. If identical titres were observed across multiple serovars (i.e., mixed reactions), the sample was classified as reactive to the all corresponding serovars.

Geoinformation mapping analysis was conducted utilizing Quantum GIS 3.16.0 software, accessible for free at www.qgis.org/ru/site/forusers/download.html. The vector layers for the borders of Ukraine's regions were downloaded from the site www.diva-gis.org/Data. Quantile classification with 5 classes of the data was chosen. With this classification, an Equal Count of oblasts fall into each class.

Information regarding the overall count of susceptible animals was extracted from statistical records provided by the State Statistics Service of Ukraine (<http://ukrstat.gov.ua>). The analysis excluded data from the occupied region of the Autonomous Republic of Crimea, the city of Sevastopol, and segments of the temporarily occupied zones within the Donetsk and Luhansk regions due to unavailability.

Results

The number of serological diagnostic samples from cattle for the detection of specific antibodies to pathogenic leptospires in Ukraine for the period 2005–2024 is shown in Figure 1.

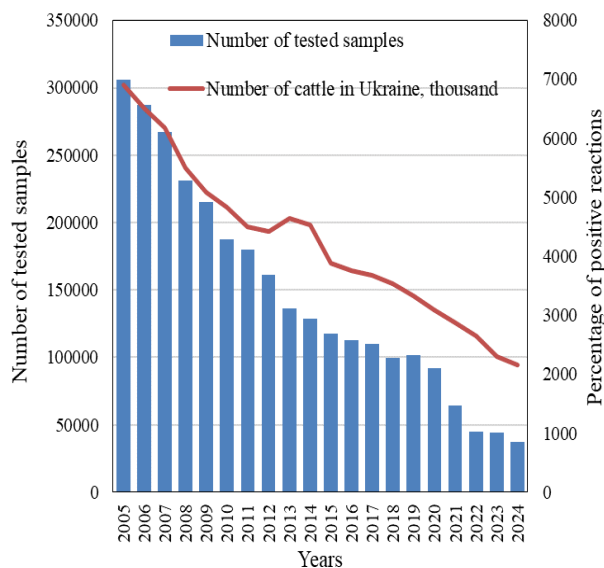


Fig. 1. Dynamics of the number of blood sera samples from cattle tested for leptospirosis and total population of cattle in Ukraine (2005–2024 years)

As can be seen from Figure 1, for the period 2005–2024, the largest number of bovine serum samples was tested in 2005, 2006 and 2007 – 305 861, 287 334 and 267 214 samples, respectively, and the smallest – in 2022, 2023 and 2024 – 45 065, 43 943 and 37 358 samples, respectively. It should be noted that during the period from 2005 to 2015, the volume of serological diagnostics of bovine leptospirosis significantly decreased from 305 861 samples in 2005 to 117 398 samples in 2015 (the number of samples tested during this period decreased by 61.6%). From 2017 to 2020, the number of samples tested was approximately the same, and from 2020 the volume of serological diagnostics began to decrease again in 2024, amounting to 37 358 studies. In general, during the analyzed period, from 2005 to 2024, the volume of serological diagnostics decreased by 8.2 times.

To compare the amount of research conducted and the number of cattle, Figure 1 shows the dynamics of the livestock population in Ukraine according to the State Statistics Service of Ukraine (data: www.ukrstat.gov.ua/operativ/operativ2006/sg/sg_rik/sg_u/tvar_u.html) as of January 1 of each year. As can be seen from the graph, the number of cattle in Ukraine also decreased during the analyzed period. Thus, in 2005–2024, the number of cattle in Ukraine decreased from 6902.9 to 2156.2 thousand head i.e., the number of cattle decreased by 68.8% during this period. In general, since Ukraine's independence, the number of cattle has decreased by 11.2 times (from 24,623.4 thousand in 1991 to 2156.2 thousand in 2024). In this case, statistics show that about 57.3% of cattle are raised (used) in the private sector.

During the period 2005–2024, veterinary laboratories of Ukraine examined 2 924 864 cattle blood serum samples, of which 148 850 were positive for leptospirosis, which is 5.1% of the total number of samples tested. The dynamics of the incidence of cattle with leptospirosis in Ukraine for the analyzed period is shown in Figure 2.

As shown in Figure 2, the seroprevalence of leptospirosis in cattle during the analyzed period was highest in 2005 and 2007 – 8.6% and 7.2%, respectively. The lowest incidence rate was observed in recent years: 2022 and 2024 – 1.8% and 1.6%, respectively. In 2009–2018, the incidence was at approximately the same level (fluctuations in seroprevalence from 4.7% to 3.6%), in the following years this figure began to decline again – from 4.2% in 2018 to 1.6% in 2024. In general, during the analyzed period from 2005 to 2024, there was a steady downward trend in the number of cattle seropositive for leptospirosis.

In the etiological structure of cattle leptospirosis in Ukraine, according to the results of studies of state laboratories of veterinary medicine, the dominant serovars are: *kabura* (serogroup *Hebdomadis*) (12.2%), *polonica* (serogroup *Sejroe*) (9.9%), *copenhageni* (serogroup *Icterohaemorrhagiae*) (5.7%), *tarassovi* (serogroup *Tarassovi*) (4.4%). Positive reactions to serovars of other leptospires were observed much less frequently: *bratislava* (serogroup *Australis*) (2.8%), *grippytyphosa* (serogroup *Grippytyphosa*) (2.2%), *pomona* (serogroup *Pomona*) (1.3%) and *canicola* (serogroup *Canicola*) (1.3%). It should be noted that during the analyzed period, a large number of positive reactions with serovars of different leptospires (mixed reactions) was observed, which accounted for 60.1% of the total number of positive animals (Fig. 3).

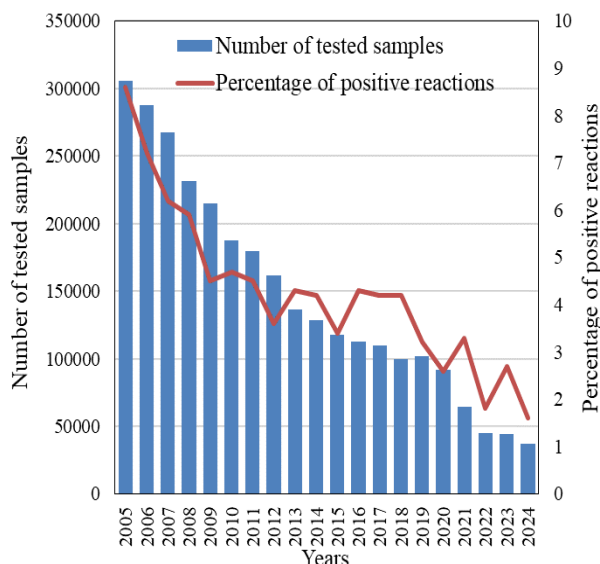


Fig. 2. Dynamics of leptospirosis infection in cattle in Ukraine (2005–2024 years)

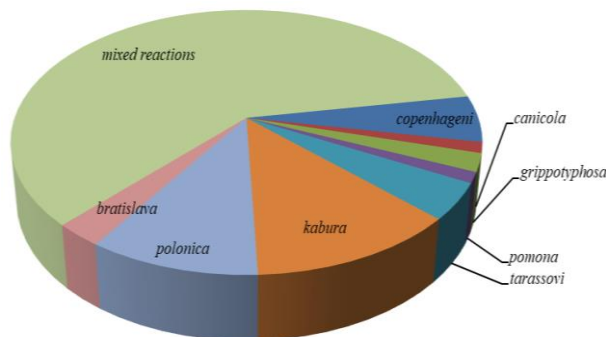


Fig. 3. Etiological structure of leptospirosis in cattle in Ukraine (2005–2024 years)

We also analyzed the dynamics of the number of seropositive cattle for the period from 2005 to 2024 to each of the eight *Leptospira* serovars used in the diagnosis of leptospirosis of this animal species in Ukraine, namely by MAT (Fig. 4).

As a result of a thorough retrospective mapping analysis of the circulation of the main diagnostic serovars among cattle in Ukraine, it was found that during the analyzed period there was a decrease in the number of positive reactions to the serovars *kabura*, *polonica* and *tarassovi*. At the same time, we established an increase in the number of reactions to the serovars *copenhageni*, *canicola* and *bratislava*. The percentage of seroprevalence of *Leptospira grippytyphosa* and *pomona* was approximately the same.

Based on the results of the analysis data, we have compiled "Maps of the distribution of cases of cattle seropositive to various leptospires serovars" for eight serogroups, which visualize the number of positive reactions of cattle sera to these serogroups in the context of Ukrainian regions using different color intensity of each region (Fig. 5).

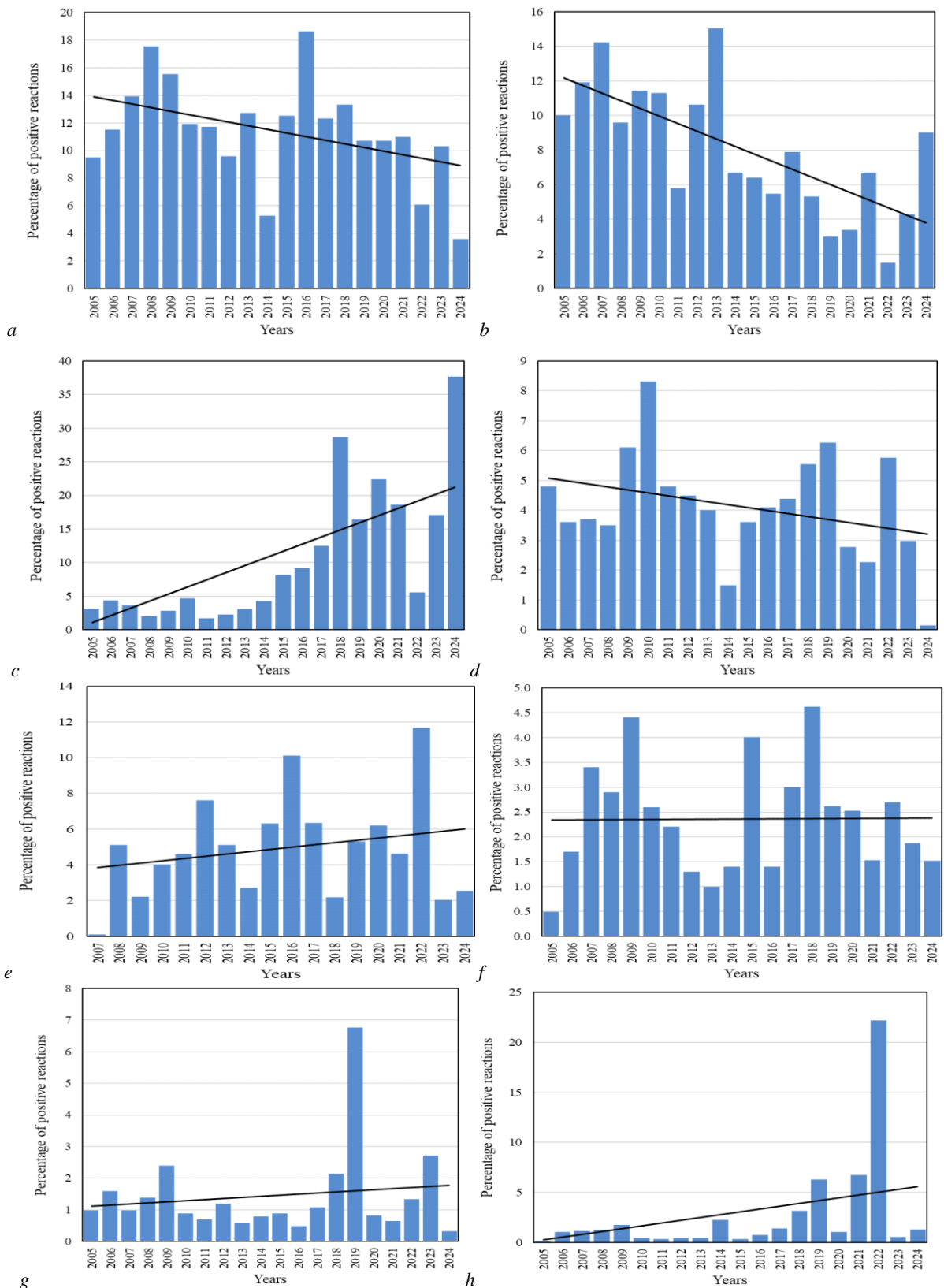


Fig. 4. Dynamics of the number of cattle seropositive to various leptospire serovars (2005–2024 years): *a* – *sv. kabura* (serogroup *Hebdomadis*); *b* – *sv. polonica* (serogroup *Sejroe*); *c* – *sv. copenhageni* (serogroup *Icterohaemorrhagiae*); *d* – *sv. tarassovi* (serogroup *Tarassovi*); *e* – *sv. bratislava* (serogroup *Australis*); *f* – *sv. grippotyphosa* (serogroup *Grippotyphosa*); *g* – *sv. pomona* (serogroup *Pomona*); *h* – *sv. canicola* (serogroup *Canicola*)

As shown on Figure 5, the highest incidence of leptospirosis in cattle is observed in the eastern and central parts of Ukraine. It has also been established that each serovar has its own peculiarities of territorial distribution in Ukraine. Thus, the serovars *polonica*, *copenhageni* and *bratislava* are most common in the eastern and central parts of Ukraine; the serovar *kabura* is most common in the eastern part of

Ukraine and Volyn region; the highest number of positive reactions to the serovars *tarassovi* and *pomona* was recorded in the northeastern part of Ukraine and in Cherkasy and Odesa regions; the serovars *grippotyphosa* and *canicola* are common in the northern and central parts of Ukraine.

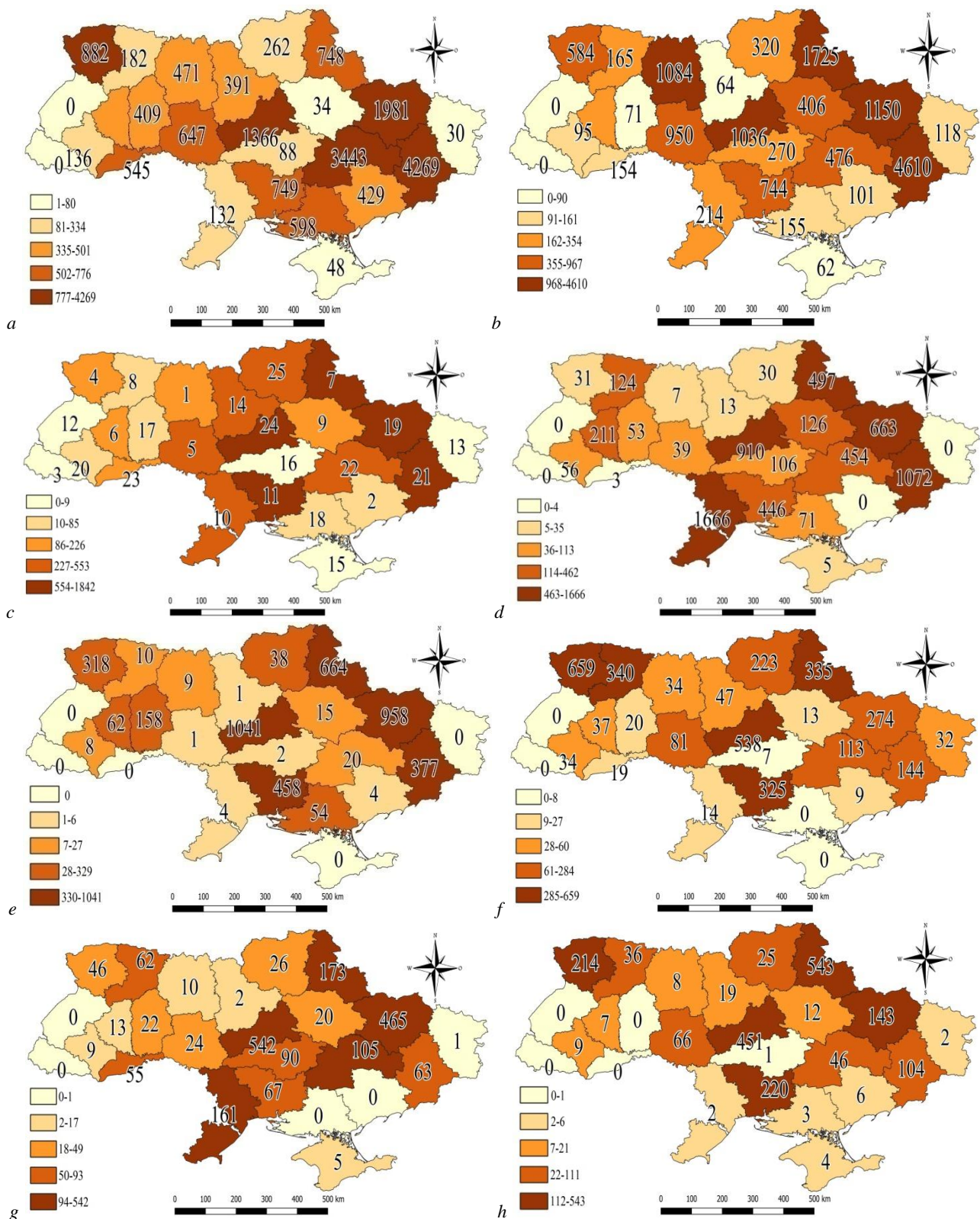


Fig. 5. Maps of the distribution of cases of cattle seropositive to various leptospire serovars (2005–2024 years):
 a – *sv. kabura* (serogroup *Hebdomadis*); b – *sv. polonica* (serogroup *Sejroe*); c – *sv. copenhageni* (serogroup *Icterohaemorrhagiae*);
 d – *sv. tarassovi* (serogroup *Tarassovi*); e – *sv. bratislava* (serogroup *Australis*); f – *sv. grippityphosa* (serogroup *Grippityphosa*);
 g – *sv. pomona* (serogroup *Pomona*); h – *sv. canicola* (serogroup *Canicola*)

Based on the studied data, a comprehensive map of the etiological composition of bovine leptospirosis in all regions of Ukraine was compiled. This map visually represents the distribution of eight different *Leptospira* serovars in the period from 2005 to 2024 using pie charts, as shown in Figure 6.

After a thorough retrospective cartographic analysis of the circulation of the main diagnostic serovars of leptospire among cattle in

Ukraine, it was found that the epizootic situation of leptospirosis of this animal species in different regions of Ukraine is heterogeneous and has its own characteristics, both in terms of the number of cases of cattle infection and the number of leptospirosis pathogens. For example, in Odesa, Volyn, Ivano-Frankivsk, Kharkiv and Chernihiv regions, cases of cattle infection with leptospire of all 8 serovars official-ly included in the diagnostic range for leptospirosis testing in Ukraine

are recorded, in this case, a significant percentage of animals were infected with several serogroups at the same time. In Zhytomyr, Luhansk and Zaporizhzhia regions, the spectrum of *Leptospira* is represented mainly by only a few serovars.

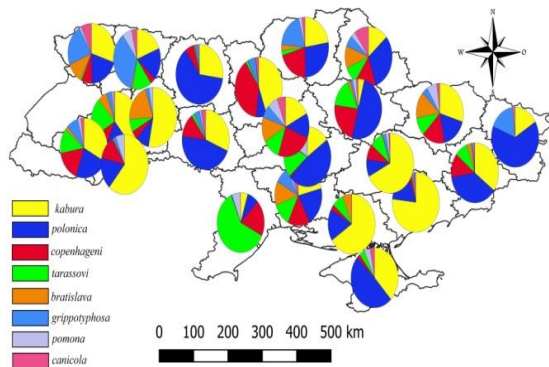


Fig. 6. Map of the etiological structure of leptospirosis of cattle in Ukraine (2005–2024 years)

Based on the analyzed data, a map of the density of leptospirosis in cattle in Ukraine over the past twenty years was compiled, which is shown in Figure 7. On this map, all regions of the country are divided into the five risk zones: very low, low, medium, high and very high.

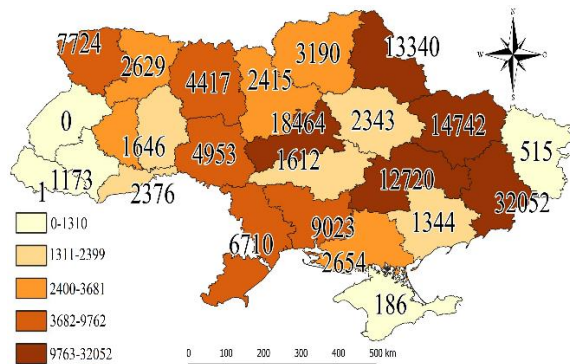


Fig. 7. Map of the density of cattle leptospirosis cases in Ukraine (2005–2024 years)

The very high-risk area includes five regions: Donetsk (32 052 positive samples), Cherkasy (18 464), Kharkiv (14 742), Sumy (13 340), Dnipropetrovsk (12 720 positive samples). The total number of positive samples for cattle leptospirosis in this zone is 91 318, which is 61.3% of the total for Ukraine. The oblasts included in this zone are characterized by a very high probability of leptospirosis in the cattle population. Ivano-Frankivsk, Luhansk, AR Crimea, Zakarpattia and Lviv oblasts have had the lowest number of leptospirosis – positive cattle over the past twenty years, and therefore are classified as a low-risk area, with a total incidence of about 1.3% (1 875 positive samples were detected).

Discussion

Thus, in the period 2005–2024, a total of 2 924 864 cattle serum samples were tested by veterinary laboratories in Ukraine. Leptospirosis was confirmed in 148 850 serum samples, corresponding to 5.1% of the total tested population. During the analyzed period, the number of tested samples decreased significantly, taking into account the year of the beginning of the analysis (2005) and the end (2024), the number of tested samples in 2024 was only 12.21% of the initial one (a reduction of 8.18 times), during this period there was a 3.2-fold decrease in the number of cattle in Ukraine (6902.9 and 2156.2 thousands head of cattle, respectively, in 2005 and 2024). The reason is that with a high density of cattle in the regions, the pathogen spreads more actively (Lau et al., 2016). We have the opposite situation, with a decrease in the number of livestock and their density.

The seroprevalence of cattle for leptospirosis during the analyzed period was highest in 2005 and 2007 – 8.6% and 7.2%, respectively.

The minimum level of seroprevalence was observed in 2022 and 2024 – 1.8% and 1.6%, respectively. In 2009–2018, the incidence was at approximately the same level (seroprevalence in the range of 3.6–4.7%), and later (2018–2024) this figure began to decline again – from 4.2% in 2018 to 1.6% in 2024. In general, during the analyzed period from 2005 to 2024, there was a steady downward trend in the number of seropositivity for bovine leptospirosis.

Our studies have shown that during the analyzed period 2005–2024 in Ukraine there was a decrease in the number of positive reactions to *Leptospira* serovars *kabura*, *polonica* and *tarassovi*, but an increase in the number of positive reactions to *copenhageni*, *canicola* and *bratislava* serovars. The percentage of seroprevalence in cattle to leptospires *grippityphosa* and *pomona* serovars remained at the same levels. According to the analysis of literature sources, this disease is a significant infectious disease of cattle with significant economic consequences worldwide, especially in Asia, the Caribbean and the African continent (Dogonyaro et al., 2020). African researchers noted that the predominant serogroups in seropositive cattle were *Sejroe* (sv. *hardjo*) (38.2%) and *Mini* (sv. *szwajizak*) (14.5%), but the rate was low for *Canicola* (sv. *canicola*) (1.8%) and *Pomona* (sv. *pomona*) (1.8%) (Dogonyaro et al., 2020). Researchers have reported that the seroprevalence of leptospires was 27.6% (Dogonyaro et al., 2020), 3.5–10.0% (Leon et al., 2008), high seroprevalence rates of leptospirosis (40.0%) in slaughtered cattle were found, for example, in slaughterhouses in Egypt (Horton et al., 2014), in St. Kitts (79.8%) (Shiokawa et al., 2019). The seroprevalence of leptospirosis in cattle varies enormously across European countries, likely reflecting local variations but also heterogeneity in design, laboratory methods and sample sizes. The risk factors positively associated with the disease are often diverse and are also related to local, environmental and climatic parameters as well as farming practices. The predominant circulating *Leptospira* serogroups in European cattle were *Sejroe* (58.5%), *Australis* (41.5%), *Grippityphosa* (41.5%), *Icterohaemorrhagiae* (37.7%) and *Pomona* (26.4%), which were found to be associated with human infections. Abortion (58.6%) and fertility disorders (24.1%) were the most commonly reported signs of leptospirosis in European cattle and generally characterized this chronic infection (Sohm et al., 2023). In Africa, the following microorganisms are isolated from animals and humans – *L. borgpetersenii*, *L. interrogans* and *L. kirschneri* (Allan et al., 2015). In Latin American countries, the incidence of this pathogen in herds is 75.0%, and the average incidence at the animal level is 44.2%. Strains of the *Sejroe* serogroup prevailed (80.3%) (Pinto et al., 2016). Differences in the prevalence of different serovars also depend on the factors such as farm contact with leptospirosis reservoirs, in particular rodents, environmental contamination, management systems and sanitation (Vinetz, 2001; Dogonyaro et al., 2020). The wide prevalence of antibodies to the *Sejroe* (sv. *hardjo*) serogroup suggests that cattle are a reservoir for this serovar and that this serovar can cause leptospirosis in cattle (Balamurugan et al., 2018). Other researchers have reported the predominance of the *Icterohaemorrhagiae* serogroup in cattle (Chadsuthi et al., 2017). The environmental impact of *Leptospira* and its epidemiology primarily determine the chronic colonization of the kidneys of reservoir animals. Cattle are recognized as a host and reservoir of serovar *hardjo* (serogroup *Sejroe*). Infection of cows with this serovar leads to chronic infections, abortions, lactation, and reduced milk yield (Sohm et al., 2023), so this disease has a significant economic impact due to the reproductive and nonreproductive losses in cattle breeding (O'Doherty et al., 2015).

The very high-risk area for leptospirosis in Ukraine includes five regions: Donetsk (32 052 positive samples during the analyzed period), Cherkasy (18 464), Kharkiv (14 742), Sumy (13 340), and Dnipropetrovsk (12 720 positive samples). The total number of positive samples for bovine leptospirosis in this zone is 91 318, which is 61.3% of the total for the country. The areas included in this zone are characterized by a very high probability of leptospirosis in the cattle population. The least risky regions in terms of bovine leptospirosis are Ivano-Frankivsk, Luhansk, Crimea, Zakarpattia and Lviv regions, as the lowest number of positive reactions to leptospirosis in this animal species was registered here (only 1.3% or 1 475 positive blood serum samples).

Cartographic analysis of the circulation of diagnostic leptospire serovars among cattle in Ukraine shows heterogeneity of the epizootic situation with regard to this disease in different regions of Ukraine. Moreover, some variants may prevail in some regions and other strains in others. In Odesa, Volyn, Ivano-Frankivsk, Kharkiv and Chernihiv regions, cases of cattle infection with leptospire of all 8 serovars officially included in the diagnostic range for leptospirosis in Ukraine are recorded, and a significant percentage of animals infected with leptospire of several serogroups at the same time is noted. In Zhytomyr, Luhansk and Zaporizhzhia regions, the spectrum of serogroups is represented mainly by only a few serovars. As epidemiological studies in other countries show, knowledge of the seroprevalence of bovine leptospirosis in a geographical region is important for veterinary practitioners and physicians, because such studies can include or exclude diseases in their differential diagnoses and reduce insufficiently substantiated and incorrect diagnoses, ultimately preventing human mortality. European researchers note that a pan-European consensus is needed on the minimum panel of serogroups that should be included in the diagnostic range, which can be further optimized with locally isolated strains (Jayasundara et al., 2021), as recommended by the World Organization for Animal Health (Petrakovsky, 2021).

The serogroups *Sejroe*, *Australis*, *Grippotyphosa*, *Icterohaemorrhagiae* and *Pomona* are the most common in cattle in Europe. Such conclusions are supported by other observations, for example, from South America (Pinto et al., 2016), Africa (Allan et al., 2015), Malaysia (Sabri et al., 2020) and New Zealand (Dreyfus et al., 2018), as well as our research. These serogroups have also been associated with human leptospirosis cases worldwide (Dupouey et al., 2014; Allan et al., 2015; Chadsuthi et al., 2017).

The diversity of pathogenic serovars in the herd significantly complicates the prevention and control of leptospirosis in such a way that the existence of several serovars in the herd leads to re-infection with some serovars after recovery from others (Orjuela et al., 2022). This can explain the significant number of mixed positive reactions with different serological variants of *Leptospira*, as they accounted for 60.1% in our studies. Our studies have shown that in the etiological structure of bovine leptospirosis in Ukraine, the dominant serovars are: *kabura* (serogroup *Hebdomadis*) (12.2%), *polonica* (serogroup *Sejroe*) (9.9%), *copenhageni* (serogroup *Icterohaemorrhagiae*) (5.7%), *tarassovi* (serogroup *Tarassovi*) (4.4%). Positive reactions to other serovars were rarely observed: *bratislava* (serogroup *Australis*) (2.8%), *grippotyphosa* (serogroup *Grippotyphosa*) (2.2%), *pomona* (serogroup *Pomona*) (1.3%) and *canicola* (serogroup *Canicola*) (1.3%). During the serological survey in Malaysia, the following serovars were detected: *hardjo-ovis*, *kabura*, *pomona*, *ballum*, *bataviae*, *javanica*, *grippotyphosa*, *autumnalis*, *sarawak*, *patoc*, *djasiman*, *shermani*, *pomona* and *polonica* (Philip & Ahmed, 2023). *Leptospira borgpetersenii* serovar *hardjo* and *L. interrogans* serovar *pomona* are the most important pathogenic serovars of cattle responsible for systemic diseases, abortion, neonatal death, weak calves and various production losses worldwide (Bolin & Alt, 2001; Wynwood et al., 2016; Regassa & Obsu, 2024).

Important risk factors for bovine leptospirosis are related to biosecurity measures and the environment. Local or even regional conditions influence the presence of *Leptospira* in the environment as well as host-bacteria interactions. Factors related to soil and water, temperature, or the composition of the environmental microbiome can determine the possibility of persistence outside the animal body (Barragan et al., 2017). Climatic conditions in Europe, including Ukraine, are becoming increasingly suitable for the survival and transmission of pathogens carried by aquatic animals and rodents, including leptospirosis (Semenza & Menne, 2009). Extreme weather conditions, exacerbated by changes in soil use (especially urbanization), increase direct and indirect contacts between leptospire, humans and animal hosts (Dupouey et al., 2014), thus increasing the risk of epizootic and epidemic outbreaks.

Bovine leptospirosis is generally underresearched, while the disease is neglected worldwide (Goarant et al., 2019), including Europe, where a limited number of countries have investigated and reported this disease. Given the veterinary and public health importance of lep-

tospirosis, as well as its economic impact, it is important to raise awareness among stakeholders, including farmers, veterinarians, and other healthcare professionals, in areas where the disease has not yet become endemic. This is especially important in Europe, where this zoonotic disease is reported in animals and humans (Sohm et al., 2023). For these reasons, diagnostic tests of blood sera for possible carriage of the pathogen are quite important elements in the system of anti-epizootic measures (Chadsuthi et al., 2022), but as we can see in Ukraine, there is a steady downward trend in the number of studies.

Leptospirosis is a significant bacterial zoonosis that is closely related to the ecological and hydrological component, and often leads to endemic, epizootic and epidemiological situations (Bharti et al., 2003; Harran et al., 2024). In many countries in rural areas, cattle are in close contact with people and are the leading source of leptospirosis pathogen for them. This is confirmed by the coincidence of serological variants isolated from animals and humans (Suwancharoen et al., 2016; Chadsuthi et al., 2017). In this case, contaminated urine of leptospirosis-carrying animals (Suwancharoen et al., 2016; Chadsuthi et al., 2018) will pose a significant risk to animals and humans, especially during floods and rains, and the pathogen can survive for many weeks and months in wet soils and surface waters (Saito et al., 2013; Chadsuthi et al., 2018). Intersectoral control measures (veterinary and medical services) include deratization, serological monitoring and sanitation of carrier animals in livestock production, control of industrial livestock wastewater, drainage of flooded areas, etc. Vaccines for specific prevention have been developed for humans and animals. However, all vaccines are serovar-specific, developed in accordance with the serovars circulating in the respective territories and livestock farms, and are not widely available. In high-risk areas, antibiotic prophylaxis may be suggested (Brett-Major & Coldren, 2012; Win et al., 2024). Climate change and warming on the planet increase the amount of precipitation and, accordingly, contribute to the survival of leptospire in such a hydrophilic environment (Andre-Fontaine et al., 2015; Ukhovskiy et al., 2022, 2023). Researchers predict that the number of outbreaks among animals and humans will only increase (Velardo et al., 2022). In order to fully characterize leptospirosis and outbreaks, epidemiological and epidemiological studies are needed that take into account changes in the environment, and all these studies should be carried out on the basis of an interdisciplinary and interdisciplinary One Health approach (Schneider et al., 2013; Pereira et al., 2018; Tsarenko et al., 2019; Munoz-Zanzi et al., 2020; Komiienko et al., 2023). In most countries and at the local level, there is still a lack of approaches and implementation of One Health in practice (Harran et al., 2024).

Conclusions

During the period 2005–2024, veterinary laboratories in Ukraine tested 2 924 864 cattle serum samples. In total, 148 850 serum samples were positive for leptospirosis, which accounted for 5.1%. The number of samples tested in 2024 was only 12.2% of the initial number (2005). The seroprevalence of leptospirosis in cattle during the analyzed period had the most significant indicators in 2005 and 2007 and amounted to 8.6% and 7.2%, respectively. The minimum level of seroprevalence was observed in 2022 and 2024 – 1.8% and 1.6%, respectively. In general, during the analyzed period from 2005 to 2024, there was a steady downward trend in the number of seropositive results for bovine leptospirosis.

Our studies have shown that during the analyzed period (twenty years) in Ukraine there was a decrease in the number of positive reactions to *Leptospira* serovars *kabura*, *polonica* and *tarassovi*, but an increase in the number of positive reactions to *copenhageni*, *canicola* and *bratislava* serovars. The percentage of seroprevalence in cattle for *grippotyphosa* and *pomona* serovars remained at the same levels.

The authors declare that there is no competing of interest.

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