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Applying One Health approach to the study of West Nile fever in the Russian Federation

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ABSTRACT

Introduction. West Nile fever is a zoonotic transmissible disease caused by flavivirus that primarily circulates in nature within an enzootic cycle between mosquitoes and birds, and causes disease cases in humans, horses, and other mammals. The rapid expansion of the West Nile fever pathogen range, development of outbreaks with severe clinical manifestations, and the lack of specific preventive tools have been the main arguments for classifying it as a potentially dangerous threat to global health. The close interconnection between human health, animal health, and ecosystems necessitates communication and coordination across the relevant sectors. One Health is an integrated, unifying approach aimed at optimizing the health of humans, animals, and ecosystems involving public health, veterinary and environment protection authorities.

Objective. Analysis of basic epizootological data on the spread of West Nile fever in the Russian Federation.

Materials and methods. The following international and Russian databases were used for literature searching: PubMed, Springer, Google Scholar, CrossRef and Russian Science Citation Index (RSCI), eLibrary, CyberLeninka, respectively. The searching was performed based on the following key words: West Nile fever, One Health, migration of birds, invertebrate hosts, control measures.

Results. In Russia, West Nile virus was first isolated in Astrakhan Oblast in 1963. Currently, presence of the pathogen has been proven in the southern and central regions of the European part of the country, in the south of Western Siberia and the Far East. The lack of disease monitoring in some Russian regions and small numbers of samples tested in most subjects of the Russian Federation hinder an objective assessment of the disease situation, so there is a need to increase the number of tests. It has been shown that the main carriers of the pathogen in our country are mosquitoes of the genera *Culex*, *Anopheles* and *Aedes*, ixodid, argasid and gamasid ticks are also involved in maintaining the virus circulation. The review describes the role of birds in the pathogen transmission, provides data on susceptibility of animals to the infection, discusses modern aspects of West Nile fever diagnosis, prevention and control.

Conclusion. Surveillance of West Nile fever presents a considerable challenge, as the virus circulates among humans, arthropods, and birds. While vaccination is an effective preventive tool, no vaccines against the disease have yet been developed in Russia. In this context, strengthening inter-authority coordination and implementing environmental control measures to limit the virus spread are essential priorities.

Keywords: review, West Nile fever, One Health, migration of birds, invertebrate hosts, control measures

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Концепция «Единое здоровье» в изучении лихорадки Западного Нила на территории Российской Федерации

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РЕЗЮМЕ

Введение. Лихорадка Западного Нила – зоонозное трансmissible заболевание, вызываемое флавивирусом, который чаще всего циркулирует в природе в энзоотическом цикле между комарами и птицами и вызывает случаи болезни у людей, лошадей и других млекопитающих. Стремительное расширение ареала возбудителя лихорадки Западного Нила, развитие вспышек с тяжелым течением болезни, отсутствие средств специфической профилактики послужили главными аргументами при отнесении его к группе потенциально опасных угроз глобальному здравоохранению. Тесная взаимосвязь между здоровьем людей, животных и экосистемой требует коммуникации и координации между соответствующими секторами. Принцип «Единое

здоровье» – это интегрированный, объединяющий подход, направленный на оптимизацию здоровья людей, животных и экосистем, включающий в себя привлечение органов общественного здравоохранения, ветеринарии и охраны окружающей среды.

Цель исследования. Анализ основных эпизоотологических данных о распространении лихорадки Западного Нила на территории Российской Федерации.

Материалы и методы. Поиск источников литературы осуществлялся с использованием международных (PubMed, Springer, Google Scholar, CrossRef) и российских (РИНЦ, eLibrary, КиберЛенинка) баз научного цитирования. Отбор материала проводился по ключевым словам: лихорадка Западного Нила, единое здоровье, миграция птиц, беспозвоночные хозяева, меры контроля.

Результаты. В России вирус Западного Нила был впервые изолирован в 1963 г. в Астраханской области. В настоящее время присутствие возбудителя доказано в южных и центральных регионах европейской части страны, на юге Западной Сибири и Дальнего Востока. Отсутствие эпизоотологического мониторинга заболевания в отдельных регионах России и низкие объемы исследуемого материала в большинстве субъектов не позволяют дать объективную оценку эпизоотической ситуации, поэтому имеется необходимость в увеличении числа проводимых исследований. Показано, что основными переносчиками возбудителя на территории нашей страны являются комары родов *Culex*, *Anopheles* и *Aedes*, также в поддержании циркуляции вируса принимают участие иксодовые, аргасовые и гамазовые клещи. В обзоре описана роль птиц в передаче возбудителя, приводятся сведения о восприимчивости животных к инфекции, рассмотрены современные аспекты диагностики, профилактики лихорадки Западного Нила и меры борьбы.

Заключение. Надзор за лихорадкой Западного Нила является достаточно сложной задачей, поскольку циркуляция вируса происходит среди людей, членистоногих и птиц. Вакцинация является эффективным средством профилактики, однако иммунологические профилактические препараты против заболевания в России пока не разработаны. В связи с этим особое внимание должно уделяться взаимодействию между различными ведомствами, а также профилактическим мерам, направленным на снижение распространения вируса в окружающей среде.

Ключевые слова: обзор, лихорадка Западного Нила, «Единое здоровье», миграция птиц, беспозвоночные хозяева, меры контроля

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INTRODUCTION

Zoonoses cause billions of human infections annually, representing a major global public health challenge [1]. The vast diversity of animal species living on the territory of the Russian Federation (RF) creates favourable conditions for the development of infectious diseases, including those transmitted by arthropods. West Nile fever (WNF), caused by a flavivirus, is a zoonotic vector-borne disease. It primarily circulates in an enzootic cycle between mosquitoes and birds, with spillover causing the disease in humans, horses, and other mammals [2, 3, 4]. West Nile virus (WNV) has been detected in more than 300 bird species, including wild, domestic, and synanthropic species. The migration routes of birds that nest across vast areas of Russia and winter in southern countries create a pathway for the introduction of the virus from wintering grounds, potentially leading to the formation of seasonal or persistent natural foci [5]. The rapid expansion of the WNV range, the emergence of severe disease outbreaks, and the absence of specific preventive measures were the primary factors justifying its classification as a potentially dangerous threat to global health – a consideration reflected in the World Health Organization's (WHO) International Health Regulations¹ [6]. The interdependence of human, animal, and ecosystem health, necessitating cooperation, communication, and coordination between the relevant sectors. One Health principle is an integrated, unifying approach aimed at sustainable balance and optimization of human, animal and ecosystem health, including the involvement of public health, veterinary and environmental protection authorities [7, 8]. The application of this approach is the key to obtaining a comprehensive understanding of WNF situation and measures for its control.

The purpose of the review is to analyse the main epizootological data on WNF spread in the RF.

MATERIALS AND METHODS

The literature sources were searched using international (PubMed, Springer, Wiley Online Library, Google Scholar, CrossRef) and Russian (RSCI, eLibrary, CyberLeninka) scientific citation databases. The selection of the material was carried out according to the keywords: West Nile fever, One Health, bird migration, invertebrate hosts, control measures.

RESULTS AND DISCUSSION

West Nile fever is caused by a flavivirus maintained in a natural transmission cycle between birds and mosquitoes. Humans, horses, and other mammals are considered incidental or dead-end hosts [9, 10]. According to the classification of the International Committee on Taxonomy of Viruses (Release 2022), WNV belongs to the genus *Ortho-flavivirus* of the *Flaviviridae* family and is a small (about 50 nm in diameter) spherical enveloped flavivirus, the genome of which consists of a single-stranded positive-sense RNA molecule encoding three structural and seven non-structural proteins [11, 12]. Based on phylogenetic analysis, WNV was grouped into 9 lineages [13]. Strains of lineages 1 and 2 are the most virulent and are capable of causing outbreaks of infection with severe neurological signs [14, 15, 16].

In 1937, WNF agent was first isolated from a woman in West Nile District in Uganda (Africa). Since then, the virus has spread widely around the world, causing outbreaks in humans and animals on all continents, including most of Africa, Eastern and Southern Europe, North America, Western Asia and the Middle East. Outbreaks in humans have been reported in South Africa, the USA, Algeria, Tunisia, Morocco, Romania, Israel, Italy, and Greece. Horse-related epizootics

¹ International Health Regulations (2005). 3rd ed. WHO; 2016. 74 p. <https://www.who.int/publications/i/item/9789241580496>



Fig. 1. Administrative territories of the Russian Federation where West Nile fever human cases were reported in 2024 (according to the data of the Volgograd Plague Control Research Institute of the Rospotrebnadzor²)

have occurred in Morocco and Italy. In Russia, direct evidence of WNV presence was first obtained in 1963 when studying Crimean-Congo hemorrhagic fever foci in Astrakhan Oblast. Southern Russian territories are historically endemic for WNV; however, global climate change has facilitated northward spread, with recent outbreaks documented in central regions and the Volga basin. Currently, the presence of the WNV agent has been proven in the southern and central regions of the European part of Russia, in the south of Western Siberia and the Far East [6, 17, 18, 19, 20].

Humans are very susceptible to WNV, but they are considered dead-end hosts. Approximately 80% of human infections with WNV are asymptomatic. In the majority of symptomatic cases (20%), patients have mild fever associated with myalgia, arthralgia, headache, fatigue, intestinal disorders, rash, and enlarged lymph nodes. Less than 1% of them develop serious neurological complications, manifested by various pathologies such as meningitis or meningoencephalitis, acute flaccid paralysis, and eye diseases. Encephalitis is the most severe neurological form, which can sometimes be fatal, especially in the elderly people and people with compromised immunity. Assessing persistent WNV infection in humans remains challenging; however, detection of the virus in urine up to 9 years post-infection demonstrates long-term renal persistence. Consequently, WNV can cause chronic human disease [11, 19].

Epidemiological surveillance of sylvatic cycle infections having transmission mechanism in our country is carried out by the Volgograd Plague Control Research Institute of the Rospotrebnadzor. There are Reference Centres for WNV, Zika fever, tick-borne infections, etc. at the Institute. According to the Reference Centre data on monitoring of WNV agent, human cases were reported in 33 Subjects of the RF in 2024 (Fig. 1). Laboratory tests of birds for WNV markers in 2024 were carried out in 34 RF Subjects, which is only 38.2% of the total number of administrative territories of the RF. Mosquitoes were tested for the infection in 76 RF Subjects (85.4% of the total number of administrative territories of the RF), ticks were tested for the infection in 58 RF Subjects (65.2%). Tests of large mammals for antibodies

to WNV were carried out in 5 RF Subjects, the virus markers detection rate was 3.4%.

The absence of epizootological monitoring for WNV in some regions of the country, combined with the low volume of samples tested in most RF Subjects, precludes an objective assessment of the disease situation. Consequently, there is a clear need to increase the volume of testing. Therewith, the exchange of data on the WNV surveillance between different authorities would provide a more complete understanding of the epidemiology and epizootology of the pathogen and improve disease prevention and control measures. To do this, it is necessary to create a single cross-departmental database based on integrated animal and environmental monitoring systems, and the data exchange mechanism should be transparent and standardized [21].

Ecology of the virus. WNV is maintained in a “bird – mosquito – bird” enzootic cycle, with birds serving as amplifying hosts. Mosquitoes become infected by feeding on the blood of infected birds and remain contagious throughout their lives. The disease agent is transmitted to animals and humans through mosquito bites (mainly of the genus *Culex*), but it can also be transmitted through organ transplantation, blood transfusion, and from mother to foetus during pregnancy [2, 22, 23]. The biological cycle of WNV spread is shown in Figure 2.

After entering the mosquito's body, the virus first infects the midgut. It then crosses the midgut barrier, disseminates via the hemolymph to other organs, and ultimately infects the salivary glands – a prerequisite for transmission to new susceptible vertebrate hosts. The virus is transmitted to humans and animals through the bites of infected mosquitoes. Once inside the host, it can replicate and lead to clinical disease [19]. At the same time, WNV does not cause apparent disease in mosquitoes [24]. The virus may also be maintained in mosquito populations via vertical

² Results of epizootological monitoring of West Nile fever in the Russian Federation in 2024 (according to the Volgograd Plague Control Research Institute of the Rospotrebnadzor). http://vniipchi.rospotrebnadzor.ru/s/203/files/directions/centre/lixoradka/analiz/145607_505.pdf (in Russ.)

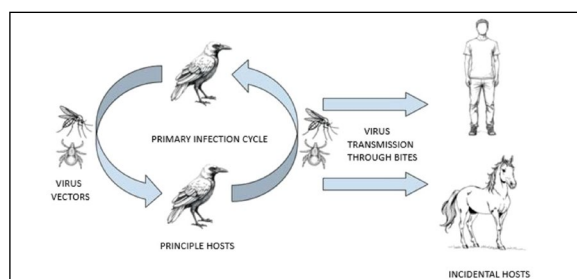


Fig. 2. Biological cycle of West Nile virus transmission (the drawing was prepared by D. A. Lunina)

transmission, in which an infected female passes the pathogen directly to her progeny. Mosquito pupae can become infected through experimental exposure to infectious mosquito secretions, potentially establishing a reservoir of infection within the mosquito population without the involvement of vertebrate hosts [25].

In humans, other vertebrates, and birds, WNV multiplies in various cell types – including keratinocytes, neutrophils, and monocytes – following the bite of an infected mosquito. The virus then disseminates via the bloodstream to peripheral organs such as liver, kidneys, and spleen. To reach the brain, the virus must cross the blood-brain barrier, which can occur in two main ways: the first involves axonal retrograde transport through the spinal cord, and the second involves transendothelial migration from the bloodstream [19].

Of approximately 100 blood-sucking mosquito species living in Russia, WNV markers have been detected in representatives of the following species: *Culex modestus* Fic., *Cx. pipiens* L. (non-autogenous form of *Cx. pipiens f. pipiens* and autogenous form of *Cx. pipiens f. molestus*), *Anopheles maculipennis* Mg., *An. claviger* Mg., *An. hyrcanus* Pall., *An. messeae* Pall., *Aedes cinereus* Mg., *Ae. geniculatus* Oliv., *Ae. vexans* Mg., *Ae. caspius* Pall., *Ae. pulchritarsis* Rond., *Ae. albopictus* Sk., *Ae. cataphylla* Dyar, *Ae. flavescens* Mull., *Ae. excrucians* Walk., *Ae. cantans* Mg., *Culiseta annulata* Schr., *Coquillettidia richiardii* Fic., *Uranotaenia unguiculata* Edw. Using Astrakhan Oblast as a model, it was shown that in anthropogenic biocenoses, the epidemiologically significant vectors include *Cx. pipiens*, *An. hyrcanus*, *Coq. richiardii*, and *An. messeae*. In natural biocenoses, *An. hyrcanus* and *Coq. richiardii* play this role. In another active WNV focus located in Volgograd Oblast, high WNV infection rates were found for mosquito species *Cx. modestus*, *Cx. pipiens*, *An. maculipennis*, and *An. hyrcanus* [6, 26]. Thus, mosquitoes of the genera *Culex*, *Anopheles* and *Aedes* are the main vectors of WNV in the RF. The genus *Culex* is ornithophilic and very aggressive towards humans. The largest number of *Culex* mosquitoes is observed in July and August. During the same period, an increase in human morbidity is observed, which is typically preceded by epizootics first among wild birds, and subsequently among domestic and synanthropic bird populations [27].

The high number of ticks in the region is also of epizootological importance. Ixodes ticks, argas ticks and gamasid mites take part in maintaining WNV circulation in Russia. Ticks of the families *Ixodidae* and *Argasidae* are believed to play a secondary role in virus transmission; their primary importance may lie in serving as overwintering reservoirs for the virus. Virus markers (antigen, RNA) were identified in entomological material and WNV isolates were recovered

from ticks of 12 species, of which most often from *Hyalomma marginatum*, *H. scupense*, *Rhipicephalus rossicus*, and *Dermacentor reticulatus* in the south of the European part of the country [6, 28]. In some years, the infection rate in *H. marginatum* ticks in anthropogenic biocenoses of the Volga Delta significantly exceeded that of mosquitoes. In addition, corvids exhibited heavy infestations with *H. marginatum* larvae and nymphs (up to 300 individuals per bird), suggesting a key role for these ticks in viral maintenance [29].

Higher ambient temperatures enhance viral replication within vectors and shorten extrinsic incubation periods, thereby promoting virus circulation and outbreak occurrence [30]. Drought reduces water flow, forming stagnant pools enriched with organic matter – optimal breeding sites for mosquitoes. Birds gather around small water bodies during droughts, and this enhances the interaction of birds and mosquitoes. Although infected mosquitoes can spread WNV over long distances – both unaided (e.g., via wind currents) and through human-mediated transport (e.g., on boats or airplanes) – infected migratory birds that carry the virus to new areas remain the primary route of WNV dissemination [31].

Role of birds in WNV transmission. Birds have the physical ability to travel thousands of kilometres across continents in a few days, crossing geographical barriers such as mountains, deserts, and seas. The intercontinental movement of wild birds across vast geographical areas facilitates the exchange of pathogens between regions and populations. Migratory birds transport various infectious agents en route and likely drive the wide-scale spread of some arboviruses. WNV circulates in nature via a sylvatic cycle involving wild birds and ornithophilic mosquitoes, alongside an urban cycle involving synanthropic and domestic birds. Migratory birds play a crucial role in WNV introduction, while resident species living in a certain territory participate in the virus amplification and local circulation. WNV persists through winter in infected female mosquitoes and avian hosts, reducing reliance on migratory birds for seasonal reintroduction [32, 33].

The most common route of infection in birds is through the bite of a mosquito that has previously fed on an infected bird. Many bird species do not develop any symptoms of the disease after infection. However, some species, such as crows, jays, and predatory birds, may die from infection. Another WNV transmission mechanism is contact. It can lead to infection, as some bird species shed large amounts of the virus with their cloacal excretions. Contact transmission can be epizootically significant when a large number of birds are concentrated in one area (in nesting colonies or at stopover sites during migration). Predatory birds such as hawks and owls, can transmit WNV via predation on infected birds and other susceptible vertebrates, which serve as either live prey or carrion. The long-term persistence of WNV in the tissues of infected animals can increase the likelihood of infection in predatory birds, even several months after the end of the mosquito season – providing a potential mechanism for the virus overwintering [34].

Five main wild bird migratory routes pass through the territory of Russia: East Atlantic, Mediterranean-Black Sea, West Asian-African, Central Asian and East Asian-Australasian³. Different populations of the same species may use different migratory routes, or they may use one. Birds

³ Krasnova E. D. About bird migrations. ROSIP. <https://birdsrussia.ru/about/articles/e-d-krasnova-o-ptichikh-pereletakh> (in Russ.)

introduce the virus into the territory of the RF from Africa, Southwest Asia, and Southeast Asia. WNV is introduced into the European part of Russia from Africa by migratory birds that follow southern and south-western flight paths in autumn. These include lake gulls, quails, swallows, ducks, sandpipers, rooks, starlings, and many other species. Birds of Western Siberia: geese, ducks, sandpipers, gulls, passerines, also follow the south-western direction during the autumn migration. Differences in avian migratory routes drive the formation of distinct WNV genotype foci in adjacent territories across Europe and Russia. Even between Volgograd and Astrakhan Oblasts, both the species composition and migratory routes of birds differ [27].

Bird migration routes in the central regions of the European part of Russia run in a meridional direction, mainly along river valleys. The bird aggregation sites are located in water-storage basins, lakes, and wetlands. Spring migration takes place between mid-March and May, while autumn migration occurs from September through October. During these periods, near-water birds – including ducks, geese, cranes, gulls, and others – undertake continuous, round-the-clock flights. More than 50 bird species have been implicated in the virus transmission in wetlands and in the vicinity of the Volga Delta. In wild birds, storks and other birds of the order *Ciconiiformes*, as well as great cormorants (*Phalacrocorax carbo*), Eurasian coots (*Fulica atra*), great cormorant (*Gallinula chloropus*), great grebes (*Podiceps cristatus*), gulls and terns (family *Laridae*) were most often identified as the virus hosts [26].

Avian species exhibit marked variation in both susceptibility to WNV infection and the clinical severity of the disease. Some avian species show virtually no symptoms of infection, but remain viremic for several days after infection, and then develop lifelong immunity. In other bird species, infection can lead to severe neurological disease and sudden death [18, 22, 35]. Most poultry species are not susceptible to the disease. In galliform birds – such as chickens, pheasants, guinea fowl, and turkeys – infection does not result in morbidity or mortality, and viremia is minimal. In contrast, predatory birds (such as hawks, owls, and eagles) appear to be susceptible to natural infection and exhibit a wide range of clinical signs [34]. In some avian species, the virus affects the central nervous system and organs such as the heart, liver, spleen, and kidneys. WNF clinical signs in birds range from nonspecific symptoms – weight loss, lethargy, and ocular disturbances – to neurological manifestations, including weakness, ataxia, head tilt, and tremors [19].

Corvids (crows, jays, magpies) serve as key sentinels for WNV circulation, as they develop fatal encephalitis – an indicator of active viral spread in emerging endemic areas. For example, in 1999, the deaths of American crows (*Corvus brachyrhynchos*) near Bronx Zoo marked the first indication of WNV introduction into the region. Since that time, these birds have been recognized as effective sentinels for the disease [36].

Susceptible animals. Some wild vertebrates, such as squirrels, chipmunks, house mice, hamsters, bats, bears, wolves, tigers, lions, striped skunks, raccoons, and crocodiles, are also affected by WNV. Wild boars serve as ideal sentinels for WNV circulation, frequently encountering infected mosquitoes in forest habitats while possessing favourable physical traits – sparse coat density and thin epidermis – that enhance mosquito feeding success. The use of alternative sentinels can help in the detection of WNV transmission routes outside of the “bird – mosquito –

bird” enzootic cycle. In particular, monitoring of wild mammals can cover a variety of habitats and time intervals during which the virus transmission may occur [37]. Among domestic animals, horses, cattle, sheep, pigs, dogs, cats, and other species are susceptible to WNV infection; however, their role in the maintenance of the virus in nature has not yet been determined [11]. As a rule, these species develop insufficient viremia for the virus transmission, and they are considered accidental or dead-end hosts [17].

Horse infections with WNV are predominantly subclinical, though some cases manifest clinical signs of the disease. Experimental infection has shown that the incubation period before onset of the first neurological signs in horses lasts for 7–9 days. The most common clinical signs are weakness, anorexia, and lethargy. Eye disorders may also develop, although blindness is considered one of the rarest clinical manifestations. Common clinical signs include enterocolitis, colic, rectal prolapse, lameness, cervical/thoracic pain, anaemia, glossitis, jaundice (suggesting hepatopathy), and dysuria (indicating nephropathy). The onset of neuroinvasive disease occurs around day 7 post-experimental infection, the clinical manifestations include ataxia, muscle weakness, fever, anorexia, lethargy, gnashing of teeth, hydrophobia, anxiety, circular movements, systemic muscle tremor and facial paralysis. WNV predominantly targets motor neurons in the brainstem (midbrain and rhombencephalon) cranial nerve nuclei, resulting in clinical signs such as dysphagia, hypersalivation, and unilateral facial nerve paralysis. In horses, the spinal cord and grey matter of the medulla oblongata and rhombencephalon are most affected, while the cerebral cortex appears to be less involved. Mortality in neuroinvasive equine cases varies from 22 to 36%; higher in older, immunocompromised horses, and foals < 12 months [38, 39].

Virus identification. WNF diagnosis plays a key role in the virus studying and disease situation monitoring. Given the risk of subclinical infection, diagnostic testing includes laboratory confirmation of the virus, as well as tests for differential diagnosis from other orthoflaviviral diseases. Accurate diagnosis requires a combination of molecular, serological, and virological methods, which provides an integrated approach to the disease detection.

Recently, WNF diagnosis has undergone significant changes with the introduction of modern molecular diagnostic methods. These techniques have significantly improved the sensitivity, specificity, and overall effectiveness of detecting this potentially dangerous infection. In particular, reverse transcription polymerase chain reaction (RT-PCR) is a molecular diagnostic tool that enables the detection of WNV RNA in blood, cerebrospinal fluid, or tissue samples during the early stages of infection, particularly in cases with severe neurological symptoms such as encephalitis or meningitis. This method is highly valuable for detecting WNV during acute-phase of the disease. However, its diagnostic window is narrow, generally limited to the first few days after infection. Real-time RT-PCR offers enhanced diagnostic capability by enabling viral load quantification, a critical parameter for monitoring of the virus circulation among humans, animals, and mosquito vectors [38]. The introduction of real-time RT-PCR for the detection of WNV RNA in urine has become an important diagnostic tool, particularly for confirming neuroinvasive WNF forms. Compared to blood testing, this approach enables detection of the virus in urine at higher concentrations and over a longer period, thereby increasing

the number of detected cases. Moreover, the use of whole blood for PCR testing proved to be more effective than using cerebrospinal fluid or serum as demonstrated during the outbreak in Arizona in 2021. This approach provides faster and more accurate diagnostic data, which is critically important for timely and effective treatment of neuroinvasive forms of the disease [40].

Enzyme-linked immunosorbent assay (ELISA) and virus neutralization tests are the most commonly used serological methods for detection of antibodies to WNV. Antibodies can be detected in horse sera with solid-phase ELISA with IgM and IgG capture, hemagglutination inhibition test (HI test), plaque reduction neutralization test (PRNT) and virus microneutralization test (MNT). IgM-ELISA detects antibodies in horse serum at clinical onset. These immunoglobulins usually develop on 7 or 10 day after infection and persist for up to 1–2 months. IgG antibodies to WNV develop shortly after the IgM response and can persist for many years. Thus, the presence of IgG without IgM is indicative of a previous infection only [11]. WNV-neutralizing antibodies are detected in horse serum two weeks after infection and can persist for more than a year. Serological assays for WNV may exhibit cross-reactivity with closely-related flaviviruses, including Usutu virus, St. Louis encephalitis virus, Japanese encephalitis virus, and tick-borne encephalitis virus. PRNT is used for differentiation of WNV from other flaviviruses.

The virus is able to replicate in susceptible cell cultures such as rabbit kidney cells (RK-13) and African green monkey kidney cells (Vero), as well as in embryonated chicken eggs. Embryonated chicken eggs or *Aedes albopictus* C6/36 cell lines can be used for the initial virus isolation with subsequent virus passaging in mammalian cells. Several passages may be required for the cytopathic effect to become apparent. Indirect immunofluorescence assay of infected cell cultures or nucleic acid detection technique are used for WNV confirmation [41].

Immunohistochemistry enables detection of macroscopic and microscopic lesions associated with West Nile encephalitis in the central nervous system tissues [42].

Treatment and prevention of the disease. The principles of WNF treatment are purely symptomatic and include maintaining adequate hydration and relieving pain associated with inflammation. Patients with severe neurological manifestations frequently require anticonvulsive therapy and respiratory assistance [34].

Global strategies for WNF prevention include two approaches: vaccination and mosquito control. Vaccines are the most effective measures against flaviviruses, especially in high-risk groups. However, despite the fact that several candidate vaccines against WNF have been developed for humans, none of them have been licensed. Currently, several vaccines for humans are undergoing phase I and II clinical trials. Development strategies include live attenuated vaccines, subunit vaccines, and recombinant DNA vaccines [43].

Vaccines for veterinary use are aimed at preventing infection of WNV-susceptible animals. Vaccines against WNF for horses have already been licensed and are available on markets of different countries. According to American scientists, four anti-WNF vaccines for horses are licensed in the United States: two inactivated whole-virion vaccines, modified live recombinant vaccine containing the canarypox virus as a vector, and a chimeric one that combines the WNV antigen with an inactivated flavivirus [44, 45]. Vaccines

against WNF have proven protective efficacy, and their use has significantly contributed to reducing the disease incidence in horses in the United States. However, despite their proven effectiveness, these vaccines still have limitations, including the need for multiple doses during primary immunization and the relatively short duration of immunity, which necessitates annual revaccination. Therefore, these aspects should be taken into account in order to improve existing vaccines and candidate vaccines under development [11]. Products intended for protection of dogs, cats, cattle, pigs, crocodiles and birds from WNV infection are under development.

To date, no safe and effective vaccine against WNF has been developed in Russia. This situation contributes to the regular occurrence of outbreaks in both endemic and non-endemic areas. Therefore, special attention should be paid to preventive measures aimed at reducing the virus spread in the environment, including among birds, animals and mosquitoes. To prevent WNV infection, individuals should take the following precautions: protect themselves against mosquito bites by using repellents, mosquito nets, and fumigators; eliminate standing water in containers around gardens and suburban areas to reduce mosquito breeding sites; avoid swampy areas, and wear protective clothing when visiting forests or the shores of water bodies. Avoid direct contact with sick or dead birds, including their faeces and feathers. When it is necessary to pick up a bird (e.g., to transport it to a veterinary centre), the persons should wear gloves and wash hands thoroughly afterward, along with any surfaces that may have been contaminated. When WNF is suspected in humans, pets, or birds, official health authorities and the state veterinary supervision service should be promptly notified [22].

CONCLUSION

West Nile virus is not classified as a candidate virus for a pandemic, as its spread and clinical significance vary depending on the region and environmental conditions. Nevertheless, it is of considerable interest to researchers in various fields, including physicians, public health workers, environmentalists, veterinarians, epidemiologists, and specialists in infectious diseases. This is due to the ability of WNV to cause severe neurological conditions in humans, especially in the elderly and immunocompromised patients, which makes it a potential threat to vulnerable population groups. In addition, clinical manifestations of WNF can be observed in animals and birds, which underlines the importance of WNV as a zoonotic pathogen. The trend for global climate change observed in recent decades contributes to the expansion of the habitat of arbovirus vectors and, consequently, increases the risk of pathogens spreading to new, previously non-endemic areas. This can lead to changes in the epidemiological context and the appearance of new foci of infection. Given these factors, WNV represents a significant concern that demands ongoing attention from the scientific community and health authorities. It can be classified to the group of potentially dangerous threats to global health, especially in the context of sustainable climate change [46].

Currently, research activities are focused on the development and improvement of new methods for WNF diagnosis, treatment and prevention. Significant emphasis is placed on molecular diagnostic techniques, as they allow for quick and reliable detection of the virus in biological samples. The immunological aspects of the disease are also being

actively studied, including the mechanism of immunity development and possible ways for effective vaccine creation. Vaccination is one of the most effective tools for disease prevention, and the development of a vaccine against WNV could be an important breakthrough for combating this infection, especially for at-risk groups. However, to date, no immunological preventive products against WNV have been developed or certified in Russia. Hence, a comprehensive, unifying One Health approach involving public health, veterinary and environmental authorities, could become the most effective and efficient mechanism to combat WNV.

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