THE RESULTS OF THE STUDY OF THE EPIDEMIOLOGICAL STATUS AND SPREAD OF DERMACENTOR RETICULATUS TICKS IN UKRAINE OVER THE LAST 10 YEARS

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Larysa Ya. Fedoniuk¹, Stepan S. Podobivskiy¹, Iryna B. Pryvrotska¹, Olena A. Miklashevska², Olga M. Marchuk¹ ¹I. HORBACHEVSKY TERNOPIL NATIONAL MEDICAL UNIVERSITY, TERNOPIL, UKRAINE ²TERNOPIL NON-COMMERCIAL ENTERPRISE "TERNOPIL COMMUNAL CITY HOSPITAL №2", TERNOPIL, UKRAINE

ABSTRACT

The aim: To analyze the study of the *D. reticulatus* ticks epidemiology and to carry out their own examinations of ticks for their infection with pathogens of infectious diseases. **Materials and methods:** Identification of ticks was performed by an optoelectronic SEO system – IMAGLAB. Detection of pathogens in the studied ticks was carried out in research laboratory of I. Horbachevsky TNMU by polymerase chain reaction (PCR) in real time using the amplifier "RotorGene – 6000".

Results: The review of scientific publications concerning an epidemiological condition of *D. reticulatus* ticks in particular countries of Europe and in Ukraine is carried out. According to the PCR results, 5 cases out of 21 samples of Borelia burgdorferi s.l. and Anaplasma phagocytophilum was detected. Researchers and students of I. Horbachevsky TNMU during 2017-2019 conducted field meetings in 74 locations from 10 regions of Ukraine: Lviv, Ivano-Frankivsk, Zakarpattia, Ternopil, Volyn, Rivne, Zhytomyr, Chernihiv, Khmelnytsky, Vinnytsia. In 2000-2018, researchers at the Department of Acarology of the I.l. Schmalhausen Institute of Zoology of the National Academy of Sciences of Ukraine the distribution of D. reticulatus in 311 locations in 79 settlements of its eastern and southern regions and the Autonomous Republic of Crimea revealed.

Conclusions: Medical geographic information system allows to create electronic cartographic models for scientific research and practical use for systematic monitoring, accounting and control of the medical and geographical situation of the study area on the prevalence of ticks and morbidity.

KEY WORDS: Ixodes ticks, Dermacentor reticulatus, distribution, epidemiology, Ukraine

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INTRODUCTION

Scientific fundamental research of Ixodes ticks in Ukraine began in the 60s of the twentieth century. In recent years, there is infectious diseases expansion associated with tick-transmitted pathogens, carriers, vectors of which are Ixodes tiks in the United States, and Europe including Ukraine is observed. For instance, tick-borne human grunocytic anaplasmosis (HGA) was first described in the United States in 1990 in Michigan, and between 1994 and 2004, 2,900 cases were reported [1]. Nowadays, Ixodes ticks in Ukraine are one of the numerous components of natural biocenoses and urbocenoses. Dermacentor reticulatus is one of the few species of Ixodes ticks that attack not only animals but also humans.

Although, there are still many unknown facts about the spreading of *D. reticulatus* in Ukraine and, in particular, its western regions [2]. Reports on the distribution of this specie, provided by a number of Ukrainian authors, is somewhat outdated.

Thus, the study of the prevalence of ticks of the genus Dermacentor reticulatus in different geographical areas, their ecological features, and biological patterns, epidemiological and medical significance remains a relevant topic for research. Based on the fact that this species of Ixodes ticks are an important vectors of the animals and human's infectious diseases pathogens transmission, the discovery of new places of its spread allows us to predict the spread of new infections in tick-occupied regions. That is why, the study of the epidemiological condition of ticks is an important aspect, which, in addition with the geographical distribution, allows us to predict the epidemiological status of different regions for tick borne infections.

THE AIM

To analyze the study of the *D. reticulatus* ticks epidemiology and to carry out their own examinations of ticks for their infection with pathogens of infectious diseases. Summarize the data of field collections of ticks of this species, conducted by scientists of I. Horbachevsky Ternopil National Medical University during 2017-2020 and researchers of the Department of Acarology of the I.I. Schmalhausen Institute of Zoology of the National Academy of Sciences of Ukraine in 2000-2018 on its distribution in the western, eastern and southern regions of Ukraine and in the Autonomous Republic of Crimea. To reveal the results of research of some aspects of biology of this species.

MATERIALS AND METHODS

The collection of ticks was carried out by authors and volunteers – students of the Medical University at their place of residence, as well as researchers of the Department of Acarology of the I.I. Schmalhausen Institute of Zoology of the National Academy of Sciences of Ukraine. It was mainly used to manually collect ticks from animals and remove ticks from grass using a "flag" method. Ticks were identified using the optoelectronic SEO system – IMAGLAB. To determine the ticks, the tables for determination given in the monographs of I.A. Akimov and E.M. Yemchuk were used.

Detection of pathogens in the studied ticks was carried out on the basis of the sector of experimental and clinical research of the interdepartmental training and research laboratory of I. Horbachevsky TNMU by the method of polymerase chain reaction (PCR) in real time using the amplifier "RotorGene-6000". To do this, prepare a suspension of each mite separately. VectorBest detection kit was used as a transport medium with cryopreservatives and as a solution for ticks preparation. In particular, "*RealBest DNA Borrelia burgdorferi senso lato*", "*RealBest DNA Borrelia Miyamotoi*", "*RealBest DNA Anaplasma phagocytophilum / Ehrlichia muris / Ehrlichia haffeensis*", "*RealBest DNA in Babesia species*", "*RealBest RNA in Babesia species*" set. A kit from the same company was used for DNA / RNA extraction.

By conducting a polymerase chain reaction was detected DNA of B. burgdorferi senso lato, B. Myamotoi, Anaplasma phagocytofilum / Erlichia muris / E. chaafeensis, Babesia sp. and tick-borne encephalitis virus (TBEV) RNA.

RESULTS AND DISCUSSION

Scientists in many countries in Europe and North America pay special attention to the study of the possibility of transmission of Ixodes ticks, including D. reticulatus, pathogens. So, Ewa J. Mierzejewska et al [3] indicate that in the eastern (5.42%) and northeastern (2.32%) regions of ticks of this species are carriers of Babesia canis. In the same regions, 52.03% and 41.75% of ticks, respectively, are carriers of Rickettsia raoulti. There are some data on the infection of D. reticulatus with spirochetes Borelia burgdorferi sl. Thus, in the western regions of the Republic of Poland, only 0.009% of ticks are carriers of these bacteria, and in the territory of the Lubelskie Voivodeship – 0.6% [4]. In France, this figure is 1.5% [5], and in the Republic of Belarus - 2,7% [6]. D. reticulatus has been shown to transmit tick-borne encephalitis viruses (TBEV). Thus, in particular, in eastern Poland, the level of infection of ticks of this species with the corresponding virus was more than 10.8% [7]. Numerous studies indicate the ability of D. reticulatus to tolerate Anaplasma phagocytophilum in many European countries, including Serbia [8].

Nowadays Ixodes ticks are one of the numerous components of natural biocenoses and urbocenoses in Ukraine, especially in its western regions. They can be observed in large quantities not only in sparse forests, on the edges, meadows and steppe areas, but also within various settlements, including large cities. Nowadays Ixodes ticks can be detected in parks, squares, recreational areas and even in small greenery along the streets or near residential buildings. This certain larva and nymph's ticks species spread can be explained by the presence of murine rodents and birds hosts, in which they are feed on; cats and dogs are excellent feeders for the adult stages of ticks [2, 9].

One of the first data on the habitats of *D. reticulatus* within Ukraine were presented in the monograph EM Yemchuk [10]. According to these data, this tick specie is found in 10 Ukraine regions (Lviv, Ternopil, Volyn, Rivne, Khmelnytsky, Vinnytsia, Kyiv, Sumy, Zhytomyr, Chernihiv), occupying mainly forest habitats.

The issue of distribution and biology of Ixodes ticks, including ticks of the genus Dermacentor in the Carpathians and Transcarpathian region was dealt with by II Turyanin [11], in which, he indicates the findings of ticks of this species in the Transcarpathian, Ivano-Frankivsk, Lviv and Chernivtsi regions.

There are some scientific sources on the distribution of *D. reticulatus* in urban areas. Thus, the monograph and scientific articles of I.A. Akimov and I.V. Nebogatkin provide data on the distribution of that species in the biotopes of Kyiv [12, 13]. According to the results of research carried out by scientists in 1988, 1991-1992, 2008-2009, the southern areas of *D. reticulatus* were established, including in the Crimea.

Regularities of attraction of this species to settlements, including cities, are revealed. G.V. Kolonin covers the study of prevalence of ticks of the genus Dermacentor in the Eastern and Western Europe in detail in the monograph in 1984 [14].

I.G. Uspenska is revealed the finding of *D. reticulatus* in the Moldavian codras (mountainous terrain) [15].

The most complete research of ticks prevalence in the Republic of Belarus, including the biology and epidemiology of pasture ticks, is discovered in the monograph E.I. Bychkova, I.A. Fedorova, M.M. Yakovich [16].

There are numerous studies of pasture ticks in the Poland. Thus, Waldemar Biaduń studied this specie within Lublin [17], Ewa J. Mierzejewska and her colleagues [3] studied *D. reticulatus* in its central regions, Anna Paziewska and colleagues [18] studied the prevalence of this specie in the northern regions of Poland, Zygner W. [19] and other scientists study the infection by Babesia species of *D. reticulatus* ticks in central Poland.

Lydia Chitimia-Dobler studied the distribution of *D. reticulatus* in Romania [20]. The biotope distribution of this specie was studied by Eva Bullová, Martin Lukáň, Michal Stanko, Branislav Petko in Slovakia [21]. Scientists Földvári G. have made an important contribution to the study of morphological features, biology, epidemiological significance and geographical distribution of *D. reticulatus* in Hungary [22, 23].

At the sector of experimental and clinical research of the interdepartmental educational-research laboratory of I. Horbachevsky TNMU, real-time PCR was used to study ticks from 21 locations using the RotorGene-6000 amplifier. In 5 cases, the fact of infection of ticks with pathogens: 3 – *Borelia burgdorferi* s.l, 1 – *Anaplasma phagocytophilum* and 1 – mixed infection of *B. burgdorferi* with *A. phagocytophilum* was detected.

Date of tick detection	Meeting place: region, district, settlement, geographical coordinates of the area	The host from which the tick was extracted	Number of ticks on one host	The pathogen detected by PCR
	Ternopil region			
5.05.2017	Pidhayetskyi district, village Lysa 49°14'32" s. w. 25°01'02" e. l.	dog	1 male, 2 females	**
5.05.2017	Ternopil district, c. lvachiv 49°38'45" n. w. 25°32'54" e. l.	cat	1 male , 1 female	*
10.09.2017.	Kremenets district, w. Roztoky 49°55'02" n. w. 25°31'24" e. l.	COW	4 males, 4 females	**
10.09.2017	Zbarazh district, c. Zbarazh 49°40'17" n. w. 25°46'15" e. l.	man	1 male	*
11.09.2017		man	1 female	*
06.10.2017		man	1 male	*
04.05.2018		man	1 female	*
04.06.2018		man	1 female	*
11.11.2018	c. Ternopil	dog	1male,	*
	49°34' n. w. 25°36' e. l.		2 females	*
16.10.2018		man	1 female	*
02 04 2019		man	1 male	Bb
14.06.2019		man	1 female	Δ
02 10 2019		man	1 female	*
02.10.2019		IIIdII	1 lemale	
6.10.2017	Ternopil district, v. Velykyy Hlybochok <u>49°37'16" n. w. 25°31'51" e.l.</u>	man	1 male	*
10.10.2017	Lanovets district, c. Lanivtsi <u>49°52'00" n. w. 26°05'14" e.l.</u>	dog	1 female	**
10.10.2017	Bereazhanskyy district, v. Zhukiv 49°30'57" n. w. 24°56'27" e.l.	man	1 female	Bb
18.10.2017	Ternopil district, v. Petryky <u>49</u> °31'40" n. w. 25°34'38" e.l.	man	1 female	*
2.11.2017	Lanovets district, v. Plyska 49°44'39" n. w. 26°05'40" e.l.	dog	2 males, 2 females	**
5.11.2017	Lanovets district, v. Molotkiv 49°49'29" n. w. 26°11'17" e.l.	cat	1 female 1 male	**
07.11.2017	Ternopil district, v. Bila 49°35'03" n. w. 25°34'41" e.l.	man	1 female	*
6.10.2018	Zboriv district, c. Pidberiztsi 49°51'26" n. w. 25°20'49" e.l.	dog	3 females	**
14.04.2019	Ternopil district. Hayi Shevchenkivski <u>49°36'05" n. w. 25°37'48" e.l.</u>	man	1 female	*
23.04.2019	Ternopil district, v. Lozova 49°36'39" n. w. 25°40'13" e.l.	dog	1 male, 2 females	**
03.05.2019	c. Shumsk <u>50°06'51" n. w. 26°06'52" e.l.</u>	dog	5 males, 1 female	**
30.09.2019	Kremenets district, v. Losyatyn 49°58'16" n. w. 25°29'23" e.l.	dog	6 female	**
29.10.2019	Pidvolochysk district, v. Skoryky <u>49°35'49" n. w. 26°08'33" e.l.</u>	cow	2 males, 7 females	**
18.11.2019	Chortkiv district, v. Palashivka <u>48°58'37" n. w. 25°34'13" e.l.</u>	dog	2 males, 5 females	**
2.12.2019	Borshchiv district, c. Borshchiv 48°48'05" n. w. 26°02'32" e.l.	cat	1 male, 2 females	**

	Table I. Distribution and biolog	av of ticks of the si	pecies Dermacento	r reticulatus (accord	ing to research b	v the laboratory	of TNMU
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	Lviv region			
29.05.2017	Busk district, м. Ostrivchyk-Pylnyy 49°53'41" n. w. 24°43'33" e.l.	horse	4 males, 8 female	Bb
10.10.2017	Zolochiv district, c. Pidhorodnye 49°47'34" n. w. 24°55'38" e.l.	dog	7 males, 3 females	**
23.04.2019	c. Socal <u>50°29' n. w. 24°17' e.l.</u>	dog	2 males, 4 females	**
3.05.2019	Yavoriv district, v. Losyno 49°56'52" n. w. 23°48'53" e.l.	cow	1 male, 6 female	**
15.09.2019	Lviv <u>49°50'30" n. w. 24°01'53" e.l.</u>	dog	2 males, 2 females	**
10.10.2019	Busky district, v. Bezbrody 49°55'06" n. w 24°33'10" e.l.	cow	8 males, 6 female	**
15.10.2019	Sokal district, v. Silets 50°17'57" n. w.24°11'53" e.l.	COW	6 female	**
1.11.2019	Zhydachiv district, v. Piddnistryany 49°25'41" n. w. 24°12'11" e.l.	dog	2 females	**
	Ivano-Frankivsk region	1		
01.04.2017	Kolomyia district, v. Runguri 48°28'41" n. w. 24°51'38" e.l.	dog	4 males	A, Bb
5.05.2017	Yaremche district, Yaremche 48°27'37" n. w. 24°33'31" e.l.	dog	1 female	*
10.05.2017	Kalush district, Kalush 49°02'39" n. w. 24°21'35" e.l.	dog	2 females	**
	Transcarpathian region	n		
14.06.2017	Mukachevo <u>48°26'29" n. w. 22°42'49" e.l.</u>	dog	1 male, 1 female	**
03.05.2019	Khust district, v. Kireshi 48°11'20" n. w. 23°21'09" e.l.	dog	1 male, 3 females	**
	Volyn region			
29.05.2017	Gorokhiv district, v. Yarivka 50°32'54" n.w. 24°54'04" e.l.	dog	7 males, 5 female	*
15.09.2017	Lyubeshiv district, v. Prochody <u>51°47'10" n.w. 25°28'11" e.l.</u>	dog	6 males, 4 females	**
19.09.2017	Lyuboml district, v. Gushcha 51°16'44" n.w. 23°45'06" e.l.	COW	6 males	**
8.10.2017	Kivertsiv district, v.Pokaschiv 50°44'51" n.w. 25°41'23" e.l.	COW	4 males, 21 female	**
10.11.2017	Gorokhiv district, v.Lobachivka 50°25'15" n.w. 24°58'05" e.l.	COW	8 males, 8 female	**
2.11.2018	Lyubeshiv district, v. Bykhiv 51°44'54" n.w. 25°16'22" e.l.	dog	4 males, 5 female	**
23.04. 2019	Lutsk district, v. Velykyy Omelyanik 50°44'19" n.w. 25°16'04" e.l.	dog	1 male	**
15.10.2019	Manevychi district, v. Kukly 51°13'28" n.w. 25°38'53" e.l.	cow	1 male, 6 females	**
18.10.2019	Volodymyr-Volynskyi district, v. Rusniv 50°44'53" n.w. 24°19'08" e.l.	COW	3 males, 2 females	**
	Rivne region			
15.04.2017	Goshchanskyi district, v. Bugryn 50°32'16" n.w. 26°31'38" e.l.	dog	4 males, 9 female	**
28.09.2017	Ostroh district, village Mezhyrich 50°18'04" n.w. 26°28'30" e.l.	man	1 female	**

12.10.17	Sarny district, v Remchycti 51°24'39" n.w. 26°30'36" e.l.	dog	11 males, 5 females	**
23.10.2017	Volodymyretskyi district, v. Svaryny 51°16'43" n.w. 26°15'16" e.l.	COW	3 males, 7 females	**
07.09.2018	Dubno <u>50°23'35" n.w. 25°44'06" e.l.</u>	cow	1 female	**
08.09.2018	Mlyniv district, v. Vovnychy 50°30'56" n.w. 25°24'19" e.l.	COW	4 males, 3 females	**
20.09.2019	Rivne <u>50°37'11" n.w. 26°15'05" e.l.</u>	cat	1 male, 1 female	**
2.10.2019.	Kostopil district, v Penjkiv 51°01'43" n.w. 26°27'24" e.l.	COW	6 males, 4 females	**
6.05.2019	Volodymyrets district, v. Bile 51°38'29" n.w. 25°59'59" e.l.	dog	3 males, 7 females	**
8.05.2019	Sarny district, Sarny 51°19'37" n.w. 26°37'59" e.l.	horse	2 males, 8 female	**
	Zhytomyr region			
13.05.2017	Korostyshiv 50°19'07" n.w. 29°03'33" e.l.	dog	2 males	*
22.05.2017	Lyubar district, v. Krasnovolitsa 50°01'56" n.w. 28°00'44" e.l.	dog	6 females	**
20.09.2018	Olevsky district, Olevsk 51°13'40" n.w. 27°38'53" e.l.	dog	2 females	**
	Khmelnytsky region			
3.10.2017	Khmelnytsky 49°25'10" n.w. 26°58'46" e.l.	dog	1 male, 15 females	**
14.10.2017	Yemilchevskyi district, t.Yemilchene 50°52'15" n.w. 27°48'26" e.l.	dog	5 males, 23 femaless	**
16.09.2017	Derazhnyansky district, t.Lozove 49°17'35" n.w. 27°17'57" e.l.	flag	7 males, 6 females	**
12.10.2017	Belogorsky district, v. Sushivcti 49°58'35" n.w. 26°21'02" e.l.	COW	7 females	**
10.10.2018	Vinkovets district, Vinkivtsi 49°01'59" n.w. 27°14'01" e.l.	dog	5 males, 4 females	**
25.10.2018	Slavutych district, Slavuta <u>50°18' n.w. 26°52' e.l.</u>	flag	1 male, 1 female	**
23.04.2019	Slavutych district, v. Golovly 50°25'25" n.w. 26°47'03" e.l.	horse	5 males, 6 females	**
26.04.2019	Yarmolynets district, v. Golohvasti 49°16'30" n.w. 26°57'46" e.l.	dog	6 female	**
	Vinnytsia region			
10.05.2017	Barsky district, c. Luka Barska <u>49°09'09" n.w. 27°47'54" e.l.</u>	COW	1 male, 8 females	**
02.05.2019	c. Illintsi <u>49°06' n.w. 29°12' e.l.</u>	dog	5 males, 8 females	**
	Chernihiv region			
06.09.2018	Nizhyn district, v. Lypiv Rig 51°04'41" n.w. 31°57'07" e.l.	dog	1 male, 5 females	**

Note: A – Anaplasma phagocytophilum; Bb – Borrelia burgdorferi s.l.;

*- no pathogens were detected; ** – PCR analysis was not performed.

An important task of the our research was not only to identify the species composition of ticks and their life forms, but also to study the biology and epidemiology of species that attack humans and animals and are able to transmit pathogens to them [24, 25]. During May 2017 – November 2019, the laboratory identified 447 specimens of ticks of the species *D. reticulatus*, including 287 females and 160 males, collected in 66 settlements from 10 regions of Ukraine.



Fig. 1. Data on the spread of D. reticulatus ticks in Ukraine (according to the results of processing in the laboratory of TNMU)



Fig. 2. Places of collection of ticks and registration of examined patients affected by ticks in the Ternopil region. Note: the examined patients are marked with circles (different colors indicate the types of employment of patients)

We recorded 74 cases ticks detection on animals, humans and in grassland, which is shown in table 1. The main feeding hosts, on which adult ticks of both sexes were found, were 4 species of animals: dogs – 34 cases, cows – 16, cats – 4, horses – 3, humans – 15 cases, in 2 cases ticks were caught on the "flag". Compared to other Ixodes ticks, in particular *Ixodes ricinus, D. reticulatus* is much less likely to attack humans, but mostly attacks animals. It was found that *D. reticulatus* attacked humans only as adult males and females (the ratio between the sexes is almost proportional), while *I. ricinus* parasitizes humans at almost all stages: larvae, nymphs, adults.

During 2000-2018, researchers of the Department of Acarology of the I.I. Schmalhausen Institute of Zoology of the National Academy of Sciences of Ukraine carried out field research to detect ticks of the species I. ricinus and D. reticulatus in the southern regions of Ukraine and in the Autonomous Republic of Crimea. In 311 points of the studied region, the ticks D. reticulatus locations were found. According to the results of research, D. reticulatus was found in 79 points of Odessa, 45 - Mykolaiv, 44 - Kherson, 60 - Zaporizhia, 57 - Donetsk regions and in 26 points of the Autonomous Republic of Crimea. It was found that the locations of D. reticulatus ticks are located in the interval: the maximum northern point is 51044'54 " north. w. (Volyn region) and the southernmost point 450 03 '05' 'north. w. (Crimea). The westernmost point within Ukraine is located at 220 42 '49' 'east. d. (Transcarpathian region) and the extreme eastern point - 380 64 '80' 'east. w. (Donetsk region).

We have created an interactive map based on the medical geographic information system, which includes all the data obtained and above (Fig. 1).

The medical geoinformation system is created as specialized medical cartographic and analytical resource designed to collect, store, accumulate, analyze and display information about the spread of tick infections. The map provides analytical information on a wide range of tick infections in order to achieve a comprehensive view of the current state of human and animal morbidity and to develop an action plan to prevent and eliminate the negative impact.

The implementation of a geographic information system allows recording, storage and display of data on the distribution of tick-borne pathogens, as well as visualize information on the spread of tick infections in a user-friendly format (spreadsheets, charts and graphs with reference to the area).

Interactive maps show the online state of the of Ixodes ticks spreading, including *D. reticulatus* in a certain area (Fig. 2). It contains the most complete information on the frequency of occurrence of different stages of development of ticks (larvae, nymphs, adults) at a certain time, on certain nutrients, their epidemiological condition et al. These maps also contain information about the infection of people with various pathogens of infectious diseases transmitted by these ticks.

The use of an interactive map provides the collection of all this data in real time using a mobile cartographic application synchronized with the geographic information system. The cartographic application installed on the researcher's smartphone will allow to carry out systematic monitoring, accounting and control over the medical-geographical situation of the studied territory in real time. Using a system of queries and intelligent search of the electronic map, it is possible to perform a spatial analysis of the prevalence of diseases caused by ticks (babesiosis, anaplasmosis, piroplasmosis, etc.) and to monitor the real threats to public health [28].

CONCLUSIONS

Ticks of the species *D. reticulatus* are most often vectors of babesia, in particular *Babesia canis*, rickettsia – *Rickettsia raoulti*, anaplasma – Anaplasma phagocytophilum, Borrelia – Borelia burgdorferi s.l. and tick-borne encephalitis. D. reticulatus is most common in humid habitats near water bodies and in urbanized biocenoses, in which murine rodents are common, and which are often visited by domestic animals and humans. The main feeding hosts for genus D. reticulatus ticks are dogs, cows, horses, cats and humans. Only adult females and males feed on humans. Medical geographic information system allows to create electronic cartographic models for scientific research and practical use for systematic monitoring, accounting and control of the medical and geographical situation of the study area on the prevalence of ticks and morbidity.

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ORCID and contributionship:

Larysa Y. Fedoniuk: 0000-0003-4910-6888 ^{A, D, E, F} Stepan S. Podobivskiy: 0000-0002-6667-1478 ^{B, C} Iryna B. Pryvrotska: 0000-0002-4610-4943 ^D Olga M. Marchuk: 0000-0002-9089-0873 ^B Olena A. Miklashevska: 0000-0002-3938-7893 ^{C, F}

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CORRESPONDING AUTHOR

Stepan S. Podobivskiy Horbachevsky Ternopil National Medical University Ministry of Health of Ukraine 1 Maidan Voli st., 46001 Ternopil, Ukraine e-mail: podobivskiy@tdmu.edu.ua

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- A Work concept and design, B Data collection and analysis, C Responsibility for statistical analysis,
- **D** Writing the article, **E** Critical review, **F** Final approval of the article