



Detection of Anti-*Borrelia burgdorferi* IgG Antibodies Among at Risk Workers in Iran; Time for Re-consideration?

Kimia Ahmadi¹, Sajede Pourmohebi², Farnaz Zahedi Avval ¹, Majid Khadem-Rezaiyan ³, Hadi Farsiani², Masoud Youssefi ^{2, 4, *}

¹Department of Clinical Biochemistry, School of Medicine, Mashhad University of Medical Sciences, Mashhad, Iran

²Department of Microbiology and Virology, School of Medicine, Mashhad University of Medical Sciences, Mashhad, Iran

³Department of Community Medicine, School of Medicine, Mashhad University of Medical Sciences, Mashhad, Iran

⁴Antimicrobial Resistance Research Center, Mashhad University of Medical Sciences, Mashhad, Iran

*Corresponding Author: Imam Reza University Hospital, Mashhad, Iran. Tel: +985138022206, Email: youssefim@mums.ac.ir

Received: 16 June, 2024; Revised: 26 November, 2024; Accepted: 12 December, 2024

Abstract

Background: Lyme borreliosis is a significant zoonotic disease with global prevalence, and due to its severe complications, early diagnosis is crucial. Limited information is available about the disease in Iran.

Objectives: Therefore, this study aimed to determine the frequency of IgG antibodies against *Borrelia burgdorferi* among high-risk personnel.

Methods: In this cross-sectional study, serum samples were collected from 91 out of 450 employees working at the main industrial slaughterhouse in Mashhad, northeast Iran. Relevant information from the participants was recorded using a pre-prepared checklist. The presence of anti-*B. burgdorferi* IgG antibodies was evaluated using the EUROIMMUN ELISA kit, following the manufacturer's instructions. The data were analyzed using SPSS software.

Results: According to ELISA results, 10 out of 91 individuals (11%) tested positive for IgG antibodies against *B. burgdorferi*. Based on optical density (OD) values, 9 participants (9.9%) were categorized as borderline, while 72 workers tested negative. The rate of positive cases was significantly higher among workers who had contact with sheep compared to those who worked with cattle (chi-square test: $P = 0.03$; OR = 8.97; 95% CI: 1.06 - 75.56).

Conclusions: Based on the ELISA technique, 11% of slaughterhouse workers tested positive for anti-*B. burgdorferi* antibodies, with the majority of these individuals working with sheep. While the possibility of cross-reactivity should be considered, this study highlights a health concern for high-risk occupations. Comprehensive diagnostic evaluations should be performed in cases of clinical suspicion.

Keywords: Lyme Disease, *Borrelia burgdorferi*, Slaughterhouse, Iran

1. Background

Lyme disease, or Lyme borreliosis, is a significant zoonotic disease caused by the spirochete *Borrelia burgdorferi* and transmitted by ticks of the *Ixodes* family. Its incidence and geographic spread have progressively increased over the past few decades (1). The primary reservoirs for *B. burgdorferi* are small mammals (2). The *B. burgdorferi* sensu lato species complex includes three genospecies: *B. burgdorferi* sensu stricto, *B. afzelii*, and *B. garinii*, which are commonly associated with Lyme disease. The distribution and symptoms of the disease vary depending on the specific bacterial species

involved. *Borrelia* species are obligate parasites with no free-living forms (3).

The life cycle of *Borrelia* alternates between two environments: Ticks and mammals or birds. Lyme *Borrelia* resides in the midgut of *Ixodes* ticks. During a blood meal, the spirochete population increases, and phenotypic changes, including the expression of outer surface protein C (OspC), occur. These changes enable the bacteria to invade the tick's salivary glands (4). The OspC expression is also critical for establishing infection in mammalian hosts (3). While tick bites are the primary mode of transmission, direct contact with infected animal tissue or blood may also rarely cause

transmission. This extracellular pathogen migrates through tissues, adheres to host cells, and evades immune clearance to cause infection (5). Although *B. burgdorferi* is typically considered an extracellular microorganism, it may adopt an intracellular form in nonphagocytic cells such as fibroblasts, potentially leading to immune escape or treatment failure (6).

Lyme disease progresses through three stages:

- Localized infection with skin manifestations.
- Disseminated infection occurring days to weeks later.
- Persistent infection, which may last for months to years.

The clinical presentation is variable. Some patients experience only localized skin infection, while others progress to later-stage manifestations, such as arthritis (3). The late-stage symptoms of Lyme borreliosis differ between the United States, Europe, and Asia. For example, an early U.S. study found that approximately 60% of untreated erythema migrans cases developed arthritis after an average of six months (7).

Lyme disease is most prevalent in the United States and Europe and is also frequently reported in Asia (8). A 2020 study by Naddaf et al. investigated the prevalence of hard ticks along Iran's Caspian Sea coast that were infected with Lyme borreliosis and relapsing fever *Borrelia*. In this study, *Ixodes ricinus* and other hard ticks were collected from various mammalian hosts, including sheep, goats, cows, camels, horses, dogs, donkeys, rodents, and poultry. Polymerase chain reaction (PCR) analysis of *Borrelia* 16S rRNA sequences revealed the presence of *Borrelia* in 71 of 501 samples from *I. ricinus* and *Rhipicephalus* ticks (9).

In endemic areas, the risk of human infection with *B. burgdorferi* is influenced by the prevalence and infestation levels of transmitting ticks and human behaviors that increase exposure. Activities like forestry work, hunting, and hiking are associated with a higher risk of infection (10). Lyme disease is highly endemic in northeastern and north-central United States, less common in central Europe, and has been reported in parts of Russia, China, Japan, Australia, India, Iran, Turkey, and North Africa. Data are limited in most African and Middle Eastern countries, and the disease appears to be rare in northern Canada and Russia. However, global warming may contribute to an increase in *I. ricinus* and *I. persulcatus* populations, altering the disease's geographic distribution (11). Iran is currently considered a non-endemic region for Lyme disease.

2. Objectives

Early diagnosis of Lyme disease is crucial to prevent severe complications. While tick bites are the primary mode of transmission, assessing infection prevalence in high-risk groups, such as slaughterhouse workers, is essential. However, there is a lack of recent regional studies on Lyme disease prevalence in such populations. Therefore, this study aimed to evaluate the prevalence of antibodies against *B. burgdorferi* in industrial slaughterhouse personnel.

3. Methods

This study is part of broader research evaluating various zoonotic infections among slaughterhouse workers in northeastern Iran. Serum sampling was conducted at the industrial slaughterhouse in Mashhad city, northeastern Iran, as previously described (12-14). Briefly, 91 samples (out of 450 workers) were included. Participants were randomly selected, and the sampling team collected blood samples over three days in the field. Required information, including demographic data and the use of personal protective equipment (PPE), was recorded in a predefined checklist. Informed consent was obtained from all participants before their inclusion in the study. The study was conducted in full compliance with the ethical principles of the Helsinki Declaration and received approval from the Ethics Committee of Mashhad University of Medical Sciences (ethical code IR.MUMS.fm.REC.1396.595).

3.1. ELISA Analysis

ELISA testing was performed on the serum samples from employees of the industrial slaughterhouse using an ELISA test kit (kit sensitivity = 96.6%, specificity = 95.2%, as stated in the kit manual). The Anti- *B. burgdorferi* VisE ELISA (IgG) kit, produced by EUROIMMUN Company (EUROIMMUN Medizinische Labordiagnostika AG, Lübeck, Germany), was used to detect IgG antibodies against *B. burgdorferi*. The ELISA procedure was carried out according to the manufacturer's instructions. This kit employs the recombinant VisE antigen of *B. burgdorferi* as the capture antigen for detecting antibodies in the serum. The VisE antigen is a surface protein of *B. burgdorferi*, featuring conserved and highly immunogenic epitopes.

3.2. Statistical Methods

Statistical analysis was conducted using SPSS software. Group comparisons were performed using chi-square or ANOVA tests. Binary logistic regression was applied to evaluate odds ratios (OR). A significance level of 0.05 was used for all calculations.

4. Results

The demographic characteristics of the study participants are summarized in Table 1. Supplementary data provide details regarding the potential for occupational contact with animal diseases and the level of adherence to health practices by the participants. The mean age was 38.71 ± 8.07 years, ranging from 23 to 58 years. According to ELISA results, 10 out of 91 individuals (11%) tested positive. Based on the kit instructions, optical density (OD) values of 9 participants (9.9%) were classified as borderline, while 72 workers were in the negative range. The ANOVA test revealed no significant relationship between serological positivity and either the age or the duration of employment of the workers ($P = 0.930$ and $P = 0.592$, respectively).

Additionally, the chi-square test showed no significant relationship between seropositivity and type of occupation, use of PPE, or direct contact with animal viscera ($P > 0.05$). However, eight individuals (19.5%) who worked with sheep tested positive, compared to only one seropositive individual (2.6%) who worked with cattle, indicating that working with sheep was significantly associated with increased seropositivity [chi-square test: $P = 0.03$; OR = 8.97; 95% CI (1.06 - 75.56)].

Table 2 presents the relationship between seropositivity and the occupational characteristics of the participants. As noted, the type of animal contact had a significant association with the antibody test response. Table 3 illustrates the relationship between the antibody test response and factors related to exposure to animal diseases and adherence to hygiene practices among participants. Although none of the evaluated factors showed a statistically significant correlation with the antibody test response ($P > 0.05$), all seropositive participants had contact with animal viscera more than once a week, though this association was not statistically significant.

5. Discussion

In this study, the prevalence of anti-*B. burgdorferi* IgG antibodies was assessed among personnel of an industrial slaughterhouse, with 91 individuals participating. The findings revealed that 11% of participants tested positive for anti-*B. burgdorferi* IgG antibodies. A 2022 systematic review and meta-analysis reported an average global prevalence of *B. burgdorferi* antibodies at 14.5% (15). In Iran, no recent reliable studies have examined the prevalence of these antibodies in the general population, limiting the ability to compare our findings with the national prevalence.

The results indicated no significant correlation between age, type of work at the slaughterhouse, and antibody test results. However, there was a notable association between the type of animal contact and antibody test responses. Among those who tested negative, 47% worked with sheep and 53% worked with cattle, whereas among those with positive antibody results, 76% worked with sheep and only 23% worked with cattle. This suggests a higher prevalence of antibodies among individuals who worked with sheep compared to those working with cattle. A 2021 study in Egypt examining 100 cattle, camels, and dogs found that while over three-quarters of the cattle were infested with ticks, no *Borrelia* cases were detected in the cattle (16).

Further analysis in our study showed that all seropositive individuals had close contact with animal viscera more than once a week, although this relationship was not statistically significant. As noted, *B. burgdorferi* transmission through contact with the blood and viscera of infected animals is rare (17).

Additionally, there was no significant relationship between the use of PPE, such as gloves, gowns, boots, and masks, and antibody test results. Similarly, adherence to hygiene practices, such as routine disinfection of hands and work tools, was not correlated with positive antibody results.

Data regarding *B. burgdorferi* in Iran is generally scarce. The detection technique is not routinely employed by clinical laboratories, which may lead to underdiagnosis of the infection. However, there are reports confirming the presence of the disease in Iran. For instance, cases have been confirmed in Isfahan, located in central Iran (18). Additional studies have identified infections in Tehran (19, 20) and Mazandaran (21). A meta-analysis has even classified Lyme disease as an emerging infection in Iran (22), with rare presentations such as neuroborreliosis also reported in the country (23). Moreover, veterinary research in Iran has focused on detecting *B. burgdorferi* in animals (24-27) and ticks (9, 28), further highlighting the potential for human infection, particularly among high-risk groups.

None of the seropositive individuals in our study reported typical Lyme migratory erythema. However, it is important to note that not all infections with *B. burgdorferi* manifest with typical erythema. Outcomes can range from asymptomatic cases (29) to non-specific arthritis (30) or neurologic manifestations (23, 31). Therefore, relying solely on the presence of a typical rash to diagnose the infection can be misleading.

The infection is prevalent across a geographical belt stretching from Asia to Europe and North America (32),

Table 1. Demographic Characteristics of Study Participants

Variables	Frequency (%)
Gender	
Male	91 (100)
Age (y)	
≤ 40	47 (51.6)
> 40	44 (48.4)
Job	
Sheep butcher	41 (45.1)
Cow butcher	38 (41.8)
Administrative	12 (13.2)
Role	
Participation in livestock slaughtering	65 (71.7)
Abattoir inspection	1 (1.3)
Transport and handling of livestock residues	9 (11.3)

Table 2. Relationship Between Demographic Characteristics of the Participants and the Positivity of Their Antibody Test ^a

Variables	Negative IgG	Positive IgG	P-Value ^b
Age (y)			0.567
≤ 40	41 (53.2)	6 (42.9)	
> 40	36 (46.8)	8 (57.1)	
Job			0.467
Butchery	66 (85.7)	13 (92.9)	
Administrative	11 (14.3)	1 (7.1)	
Type of livestock			0.048
Sheep	31 (47)	10 (76.9)	
Cow	35 (53)	3 (23.1)	
Type of work			0.262
Slaughtering of livestock or livestock visceral disposal	61 (91)	13 (100)	
Other	6 (9)	0 (0)	

^a Values are expressed as No. (%).

^b The chi-square test has been used to compare between two groups.

including the Middle East (32, 33). However, its frequency may be underestimated due to the unavailability of standard serological diagnostic facilities.

In a 2020 study, Obaidat et al. examined the prevalence of anti- *B. burgdorferi* antibodies in the Jordanian population. Serum samples from 824 healthy individuals from various regions of Jordan were collected and tested. The results indicated that 11.7% of participants were positive for these antibodies (34). Similarly, Brummitt et al., in a study published in 2020, investigated the prevalence of anti- *B. burgdorferi* and *Borrelia miyamotoi* antibodies in blood donors in California, USA. They analyzed 1,700 blood samples using ELISA and confirmed positive cases with a Western

blot test. The findings showed that only 0.47% of individuals tested positive for anti- *B. burgdorferi* antibodies (35).

The prevalence of anti- *B. burgdorferi* antibodies in our study (11%) was significantly higher than the prevalence reported by Brummitt et al. (35). This difference may be attributed to variations in geographic regions, technical methodologies, and study populations. Blood donors typically represent the general population, while our study group comprised slaughterhouse personnel, a high-risk population with frequent exposure to animals, increasing their likelihood of exposure to the pathogen.

Our study holds important epidemiological and clinical implications. Future research should focus on

Table 3. Characteristics Related to the Possibility of Contact with Animal Diseases and the Level of Adherence to Hygiene, Principles by the Participants ^a

Properties and Times	Negative IgG	Positive IgG	P-Value
Contact with animal viscera			0.240 ^b
More than 1 time per week	70 (90.9)	14 (100)	
Less than 1 time per week	7 (9.1)	0 (0)	
History of hand cutting over a year			0.154 ^b
> 5 times	48 (63.2)	6 (42.9)	
≤ 5 times	28 (36.8)	8 (57.1)	
External parasite infestation over a year			0.984 ^b
> 5 times	27 (36)	5 (35.7)	
≤ 5 times	48 (64)	9 (64.3)	
Use of mask			0.335 ^b
Always	28 (36.4)	7 (50)	
Sometimes, seldom, never	49 (63.6)	7 (50)	
Use of gloves			> 0.999 ^c
Always	72 (96)	14 (100)	
Sometimes, seldom, never	3 (4)	0 (0)	
Use of gowns and aprons			> 0.999 ^c
Yes	73 (94.8)	14 (100)	
No	4 (5.2)	0 (0)	
Use of work boots			> 0.999 ^c
Yes	73 (94.8)	14 (100)	
No	4 (5.2)	0 (0)	
The amount of use of PPE (including masks, gloves, gowns and boots)			0.335 ^b
Complete (use of all 4 items)	28 (36.4)	7 (50)	
Relative (use 3 or less)	49 (63.6)	7 (50)	
Disinfection rate of work tools			0.784 ^b
Always	9 (11.7)	2 (14.3)	
Seldom	68 (88.3)	12 (85.7)	
Hand and face disinfection rate			0.406 ^b
Always	10 (13)	3 (21.4)	
Seldom	67 (87)	11 (78.6)	

Abbreviation: PPE, personal protective equipment.

^a Values are expressed as No. (%).

^b The chi-square test has been used to compare between two groups.

^c The Fisher's exact test has been used to compare between two groups.

investigating the prevalence of *Borrelia* antibodies in diverse populations. A limitation of our study was the relatively small sample size and the absence of data on the general population, such as blood donors. Additionally, the study relied solely on ELISA for laboratory analysis without confirmatory testing using the Western blot technique. While the ELISA kit manual indicated high specificity, the potential for cross-reactivity cannot be ruled out (36). Therefore, this study serves as a preliminary investigation and highlights the need for further research.

Despite these limitations, our study's strength lies in being the first to examine anti- *B. burgdorferi* IgG

antibodies in slaughterhouse workers in Iran. Future studies should explore the prevalence of anti-*Borrelia* antibodies in the general population, including blood donors and other high-risk groups. Moreover, periodic studies should be conducted to monitor trends and changes in the prevalence of the disease in both general and high-risk populations.

5.1. Conclusions

Approximately 11% of slaughterhouse workers tested positive for *B. burgdorferi* antibodies, with the majority of these individuals frequently handling sheep. All individuals with positive antibody tests had contact

with animal viscera more than once per week; however, this association was not statistically significant. Based on our literature review, no study to date has investigated the prevalence of *B. burgdorferi* antibodies in the Iranian population. The present study serves as a health warning for high-risk occupations, emphasizing the need for full diagnostic evaluations in cases of clinical suspicion.

Acknowledgements

The authors are grateful to the staff of the main industrial slaughterhouse of Mashhad. This work was supported by the vice president of research at Mashhad University of Medical Sciences (grant no: 961095).

Supplementary Material

Supplementary material(s) is available [here](#) [To read supplementary materials, please refer to the journal website and open PDF/HTML].

Footnotes

Authors' Contribution: Study concept and design: M. Y.; Acquisition of data: S. P. and F. Z. A.; Analysis and interpretation of data: M. Y. and H. F.; Drafting of the manuscript: K. A. and S. P.; Critical revision of the manuscript for important intellectual content: M. Y. and H. F.; Statistical analysis: M. K. R.; Administrative, technical and material support: S. P., K. A. and F. Z. A.; Study supervision: M. Y.

Conflict of Interests Statement: The authors declared no conflict of interests.

Data Availability: The dataset presented in the study is available on request from the corresponding author during submission or after publication.

Ethical Approval: The study was conducted in full compliance with the Helsinki Declaration's ethical principles and received approval from the Ethics Committee of the Mashhad University of Medical Sciences (ethical code: IR.MUMS.fm.REC.1396.595).

Funding/Support: This work was supported by vice president of research Mashhad University of medical sciences (grant no: 961095).

Informed Consent: Informed consent was obtained from all participants before their inclusion in the study.

References

1. Kalish RA, McHugh G, Granquist J, Shea B, Ruthazer R, Steere AC. Persistence of immunoglobulin M or immunoglobulin G antibody responses to *Borrelia burgdorferi* 10-20 years after active Lyme disease. *Clin Infect Dis*. 2001;**33**(6):780-5. [PubMed ID: [11512082](#)]. <https://doi.org/10.1086/322669>.
2. Barbour AG, Duong JV, Long AD. Lyme Disease Agent Reservoirs *Peromyscus leucopus* and *P. maniculatus* Have Natively Inactivated Genes for the High-Affinity Immunoglobulin Gamma Fc Receptor 1 (CD64). *Pathogens*. 2023;**12**(8). [PubMed ID: [37624016](#)]. [PubMed Central ID: [PMC10458454](#)]. <https://doi.org/10.3390/pathogens12081056>.
3. Steere AC, Coburn J, Glickstein L. The emergence of Lyme disease. *J Clin Invest*. 2004;**113**(8):1093-101. [PubMed ID: [15085185](#)]. [PubMed Central ID: [PMC385417](#)]. <https://doi.org/10.1172/JCI21681>.
4. Stanek G, Wormser GP, Gray J, Strle F. Lyme borreliosis. *Lancet*. 2012;**379**(9814):461-73. [PubMed ID: [21903253](#)]. [https://doi.org/10.1016/S0140-6736\(11\)60103-7](https://doi.org/10.1016/S0140-6736(11)60103-7).
5. Xu G, Fang QQ, Keirans JE, Durden LA. Molecular phylogenetic analyses indicate that the Ixodes ricinus complex is a paraphyletic group. *J Parasitol*. 2003;**89**(3):452-7. [PubMed ID: [12880241](#)]. [https://doi.org/10.1645/0022-3395\(2003\)089\[0452:MPAITT\]2.0.CO;2](https://doi.org/10.1645/0022-3395(2003)089[0452:MPAITT]2.0.CO;2).
6. Rahn DW, Malawista SE. Lyme disease: recommendations for diagnosis and treatment. *Ann Intern Med*. 1991;**114**(6):472-81. [PubMed ID: [1994795](#)]. <https://doi.org/10.7326/0003-4819-114-6-472>.
7. Steere AC, Schoen RT, Taylor E. The clinical evolution of Lyme arthritis. *Ann Intern Med*. 1987;**107**(5):725-31. [PubMed ID: [3662285](#)]. <https://doi.org/10.7326/0003-4819-107-5-725>.
8. Gray J, Kahl O. Tick Ecology and the Eco-Epidemiology of *Borrelia burgdorferi* sensu lato. In: Hunfeld K, Gray J, editors. *Lyme Borreliosis*. Berlin, Germany: Springer; 2022. p. 31-45. https://doi.org/10.1007/978-3-030-93680-8_2.
9. Naddaf SR, Mahmoudi A, Ghasemi A, Rohani M, Mohammadi A, Ziapour SP, et al. Infection of hard ticks in the Caspian Sea littoral of Iran with Lyme borreliosis and relapsing fever borreliae. *Ticks Tick Borne Dis*. 2020;**11**(6):101500. [PubMed ID: [32993956](#)]. <https://doi.org/10.1016/j.ttbdis.2020.101500>.
10. Kugeler KJ, Farley GM, Forrester JD, Mead PS. Geographic Distribution and Expansion of Human Lyme Disease, United States. *Emerg Infect Dis*. 2015;**21**(8):1455-7. [PubMed ID: [26196670](#)]. [PubMed Central ID: [PMC4517724](#)]. <https://doi.org/10.3201/eid2108.141878>.
11. Ozdenorel E. GIS and Remote Sensing Use in the Exploration of Lyme Disease Epidemiology. *Int J Environ Res Public Health*. 2015;**12**(12):15182-203. [PubMed ID: [26633445](#)]. [PubMed Central ID: [PMC4690907](#)]. <https://doi.org/10.3390/ijerph121214971>.
12. Khadem-Rezaian M, Azari Garmjan GA, Jarahi L, Ghazvini K, Youssefi M. Seroprevalence of Q Fever and Risk Factors Affecting Transmission of *Coxiella burnetii* in Industrial Slaughterhouse; A Survey from Northeastern Iran. *Health Scope*. 2023;**12**(1). <https://doi.org/10.5812/jhealthscope-132858>.
13. Youssefi M, Khadem-Rezaian M, Azari-Garmjan GA, Jarahi L, Shamsian AA, Moghaddas E. Prevalence of Toxoplasma and Echinococcus IgG antibodies in slaughterhouse workers, a serosurvey in Northeast Iran. *Ann Parasitol*. 2018;**64**(4):391-7. [PubMed ID: [30738424](#)]. <https://doi.org/10.17420/ap6404.176>.
14. Shahhosseini N, Azari - Garmjan GA, Khadem Rezaian M, Haeri A, Nowotny N, Fooks AR, et al. Factors Affecting Transmission of Crimean - Congo Hemorrhagic Fever among Slaughterhouse Employees: A Serosurvey in Mashhad, Iran. *Jundishapur J Microbiol*. 2018;**11**(3). <https://doi.org/10.5812/jjm.57980>.
15. Dong Y, Zhou G, Cao W, Xu X, Zhang Y, Ji Z, et al. Global seroprevalence and sociodemographic characteristics of *Borrelia burgdorferi* sensu

- lato in human populations: a systematic review and meta-analysis. *BMJ Glob Health*. 2022;**7**(6). [PubMed ID: 35697507]. [PubMed Central ID: PMC9185477]. <https://doi.org/10.1136/bmjgh-2021-007744>.
16. Elhelw R, Elhariri M, Hamza D, Abuowarda M, Ismael E, Farag H. Evidence of the presence of *Borrelia burgdorferi* in dogs and associated ticks in Egypt. *BMC Vet Res*. 2021;**17**(1):49. [PubMed ID: 33494772]. [PubMed Central ID: PMC7830850]. <https://doi.org/10.1186/s12917-020-02733-5>.
 17. Barbour AG. Laboratory aspects of Lyme borreliosis. *Clin Microbiol Rev*. 1988;**1**(4):399-414. [PubMed ID: 3069200]. [PubMed Central ID: PMC358062]. <https://doi.org/10.1128/CMR.1.4.399>.
 18. Emami Namini AR, Fatemi Naeini F, Ghorbani A. [Case report of Lyme disease in Isfahan]. *J Isfahan Med Sch*. 2005;**23**(76-77):31-3. FA.
 19. Tabatabaie P, Siadati A. A case of Lyme disease (Lyme borreliosis). *Acta Med Iran*. 2006;**44**(3).
 20. Chams-Davatchi C. The first endemic case of Lyme Borreliosis in Iran. *Med J Islam Repub Iran*. 1997;**11**(3):237-9.
 21. Adabi M, Firoozjahi AR, Ghasemi M. [Report of a case of Lyme disease in Mazandaran]. *Iran J Dermatol*. 2004;**8**(suppl):21-5. FA.
 22. Khoobdel M, Jafari AS, Telmadarraiy Z, Sedaghat MM, Bakhshi H. Tick-borne pathogens in Iran. *Asian Pac J Trop Med*. 2021;**14**(11):486-504. <https://doi.org/10.4103/1995-7645.329009>.
 23. Sayad B, Babazadeh A, Barary M, Hosseinzadeh R, Ebrahimpour S, Afshar ZM. Lyme neuroborreliosis: A case report. *Clin Case Rep*. 2023;**11**(8). e7702. [PubMed ID: 37554577]. [PubMed Central ID: PMC10405229]. <https://doi.org/10.1002/ccr3.7702>.
 24. Mosallanejad B, Avizeh R, Razi Jalali MH, Pourmahdi M. A serological survey on *Borrelia burgdorferi* infection among companion dogs in Ahvaz district, southwestern Iran. *Comp Clin Pathol*. 2015;**24**(6):1559-63. <https://doi.org/10.1007/s00580-015-2116-x>.
 25. Hanifeh M, Malmasi A, Virtala AMK, Nikbakht-Brujeni GR, Zahraei Salehi T, Rahbari S. Seroprevalence, geographic distribution and risk factor analysis of *Borrelia burgdorferi* sensu lato in naturally exposed dogs of Iran. *Afr J Microbiol Res*. 2012;**6**(25):5353-61. <https://doi.org/10.5897/ajmr12.361>.
 26. Zarei Chaleshtory M, Keihani P, Momtaz H, Hamze Ali Tehrani M, Hosseini SR. [Prevalence of *Borrelia burgdorferi* in Guard Dogs in Isfahan, Iran]. *J Vet Res*. 2023;**78**(3). FA.
 27. Esmailnejad A, Tabatabaie M, Abbaszadeh Hasiri M, Sheikhi F. Serological evidence of Borreliosis among companion dogs in Fars Province, South of Iran. *Serological evidence of Borreliosis among companion dogs in Fars Province, South of Iran. J Zoo Dis*. 2017;**2**(1):1-8.
 28. Enferadi A, Ownagh A, Tavassoli M. Molecular Detection of *Borrelia* spp. in Ticks of Sheep and Goats by Nested PCR Method in West Azerbaijan Province, Iran. *Vector Borne Zoonotic Dis*. 2023;**23**(12):605-14. [PubMed ID: 37722020]. <https://doi.org/10.1089/vbz.2023.0039>.
 29. Carlsson H, Ekerfelt C, Henningsson AJ, Brudin L, Tjernberg I. Subclinical Lyme borreliosis is common in south-eastern Sweden and may be distinguished from Lyme neuroborreliosis by sex, age and specific immune marker patterns. *Ticks Tick Borne Dis*. 2018;**9**(3):742-8. [PubMed ID: 29502989]. <https://doi.org/10.1016/j.ttbdis.2018.02.011>.
 30. Zangeneh M, Haghighi A, Asgari N. [Frequency of Lyme arthritis in patients with unknown subacute arthritis]. *Med Sci J Islam Azad Univ*. 2012;**21**(4):305-10. FA.
 31. Murray TS, Shapiro ED. Lyme disease. *Clin Lab Med*. 2010;**30**(1):311-28. [PubMed ID: 20513553]. [PubMed Central ID: PMC3652387]. <https://doi.org/10.1016/j.cll.2010.01.003>.
 32. Perveen N, Muzaffar SB, Al-Deeb MA. Ticks and Tick-Borne Diseases of Livestock in the Middle East and North Africa: A Review. *Insects*. 2021;**12**(1). [PubMed ID: 33477991]. [PubMed Central ID: PMC7835866]. <https://doi.org/10.3390/insects12010083>.
 33. Onal U, Aytac Erdem H, Uyan Onal A, Resat Sipahi O. Systematic review of Lyme disease in Turkey. *Trop Doct*. 2019;**49**(3):165-70. [PubMed ID: 31018773]. <https://doi.org/10.1177/0049475519843387>.
 34. Obaidat MM, Alshehabat MA, Hayajneh WA, Roess AA. Seroprevalence, spatial distribution and risk factors of *Borrelia burgdorferi* sensu lato in Jordan. *Comp Immunol Microbiol Infect Dis*. 2020;**73**:101559. [PubMed ID: 33086189]. <https://doi.org/10.1016/j.cimid.2020.101559>.
 35. Brummitt SI, Kjemtrup AM, Harvey DJ, Petersen JM, Sexton C, Replogle A, et al. *Borrelia burgdorferi* and *Borrelia miyamotoi* seroprevalence in California blood donors. *PLoS One*. 2020;**15**(12). e0243950. [PubMed ID: 33370341]. [PubMed Central ID: PMC7769429]. <https://doi.org/10.1371/journal.pone.0243950>.
 36. Grazlewska W, Holec-Gasior L. Antibody Cross-Reactivity in Serodiagnosis of Lyme Disease. *Antibodies*. 2023;**12**(4). [PubMed ID: 37873860]. [PubMed Central ID: PMC10594444]. <https://doi.org/10.3390/antib12040063>.