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Diagnosis and Treatment of *Anaplasma* Sp. in Goats: Clinical, Microscopic, and Molecular Approaches

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Abstract

Anaplasmosis is a tick-borne rickettsial disease affecting domestic and wild animals, particularly in tropical and subtropical regions worldwide. Over the years, several species of Anaplasma have been identified. Although numerous studies have documented their prevalence and distribution, gaps remain in our understanding of their pathogenic mechanisms and optimal treatment strategies, particularly in goats. This study aimed to identify *Anaplasma* species using both microscopic and molecular diagnostic techniques. A treatment regimen consisting of oxytetracycline and enrofloxacin was administered to infected goats. Following the implementation of this protocol, a significant reduction in mortality within the herd was observed, leading to the cessation of deaths in a relatively short period. This study underscores the effectiveness of the proposed treatment and contributes to the understanding of anaplasmosis management in goats.

Keywords: Anaplasmosis, Goat, Oxytetracycline, Enrofloxacin, Anemia

Introduction

Tick-borne diseases have significantly increased over the past 30 years, particularly in tropical and subtropical regions, posing a major threat to livestock health and productivity. Among these, *Anaplasma* spp. infections in small ruminants cause severe economic losses due to reduced productivity, increased morbidity, and mortality. The widespread adoption of molecular diagnostics, with their enhanced sensitivity and reliability, has improved the accurate detection of these pathogens in livestock. Ticks are major vectors of bacterial and protozoal infections affecting farm animals, including anaplasmosis, which remains a significant concern in goat herds(1-4).

Anaplasma species have been identified as causative agents of diseases in a wide range of domestic and wild animals on a global scale (1,5,6). Notably, certain species, including *A. phagocytophilum*, *A. ovis*, *A. capra*, and *A. platys*, have been demonstrated to possess zoonotic potential, underscoring highlighting their significance in public health(5,6,7-10).

These rickettsia are capable of affecting various cell types, including monocytes, neutrophils, erythrocytes, and platelets, depending on the host species(2,11). *Anaplasma phagocytophilum* primarily targets neutrophils and eosinophils in both infected animals and humans, while *A. marginale*, *A. centrale*, and *A. ovis* predominantly affect the erythrocytes of ruminants, and *A. bovis* targets bovine monocytes. *A. platys* has been observed to impact canine platelets; however, *A. capra* has been shown to affect endothelial cells, and its detection in blood smears remains questionable(2,5,8,11,12).

The present study aimed to investigate the presence of *Anaplasma* spp. in a goat herd in Turkey exhibiting clinical signs consistent with anaplasmosis through clinical evaluations, laboratory analyses, and molecular testing.

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Moreover, the objective was to evaluate, for the first time, the therapeutic efficacy of a combination of oxytetracycline and enrofloxacin in the treatment of goats affected by anaplasmosis.

Material and Methods

Animals and Herd Anamnesis

Firstly, the animal material consisted of two goats from an extensive family farm in Çanakkale, which were brought to Bursa Uludağ University (BUU) Veterinary Faculty Animal Hospital Subsequently, 22 goats from the same flock were included in the study. The herd consisted of 190 Saanen and Saanen crossbred goats. This research was conducted with the ethical approval of the Committee of Animal Experimentation of BUU University (Bursa Uludağ University-2020-06/90). The herd anamnesis revealed that the herd had been affected by a disease for the past month, despite treatment with various medications, including imidocarb, oxytetracycline LA, tulathromycin, and doramectin. This treatment had no success in halting the disease, which resulted in the death of 35 goats. The herd owner reported that the disease was characterized by high fever, and some of the deceased animals displayed icterus; however, none showed signs of hemoglobinuria.

The samples collected from the herd were subjected to testing for *C. perfringens* (ELISA), infectious necrotic hepatitis (FAT), goat liver pain, and sheep-goat plague (molecular techniques). However, the results of these tests were negative. It was reported that the animals were primarily raised on pasture and were also given pellet milk feed.

Two goats brought to our faculty were found to have varying degrees of anemia and pneumonia. A detailed examination of blood smears revealed the presence of inclusions within neutrophils, suggesting the existence of *Anaplasma* spp. Based on the clinical and laboratory findings, the disease was suspected to be consistent with *Anaplasma* spp. infection. Therefore, the objective was to elaborate the diagnostic process in the herd through clinical, hematological, and molecular diagnostic methods, followed by the development of a treatment plan.

Sample Collection and Laboratory Analysis

In this study, a total of 24 goats were selected for analysis, including two goats that were brought to our hospital and 22 out of 100 diseased goats from the flock that were found to be diseased. Blood samples were collected from each animal, with two millilitres of blood with EDTA and five millilitres of anticoagulant-free blood being obtained. The white blood cell (WBC) count, packed cell volume (PCV), and platelet (PLT) levels were then measured using conventional methods. Furthermore, the activities of aspartate aminotransferase (AST) and gamma-glutamyl transferase (GGT) in the blood serum of the two goats initially brought to our faculty were determined using a dry chemistry analyzer (Ref: 10745081; Reflotron Plus, Roche Diagnostic GmbH, Germany). Additionally, the lungs of the necropsied goat were sent to the laboratory for bacterial culture.

The microscopic evaluation

Blood smears were prepared and stained with Diff-Quick stain. The presence of *Anaplasma* species in erythrocytes and granulocytes was evaluated by examining the samples at 100x magnification using a light microscope. As a result of parasitological examinations, the presence of *A. phagocytophilum* or an *A. capra*-like morula in granulocytes was observed under the microscope, prompting the use of molecular diagnostic methods.

DNA extraction, PCR amplification, and sequencing

The total genomic DNA (gDNA) was extracted using the GeneJET Genomic DNA Purification Kit (Thermo Scientific, Waltham, MA, USA) in accordance with the manufacturer's protocol. The final nucleic acid was eluted in 50 µL of eluent buffer and stored at -20 °C. The DNA concentrations of the samples were then measured using Qubit 3.0 Fluorometer Quantitation (Thermo Scientific, Waltham, MA, USA) in order to standardise the amount of gDNA used in the subsequent PCR amplifications.

The gDNAs extracted from blood samples were then subjected to a series of PCR analyses utilizing the primers EHR16SD (5'-GGTACCYACAGAAGAAGTCC-3') and EHR16SR (5'-TAGCACTCATCGTTTACAGC-3') which were designed to amplify a ~345 base pair (bp) fragment of the 16S ribosomal RNA (16S rRNA) gene(13). The amplifications were carried out in a total volume of 25 µL, including 12.5 µL of commercial ready to use mastermix (DreamTaq Green PCR Master Mix, Thermo Fisher Scientific, Waltham, MA, USA), 1 µM of each primer and 10 to 30 ng of gDNA. The reactions were performed in a C1000 Touch™ Thermal Cycler (Bio-Rad, Hercules, CA, USA), with the cycling parameters described by Inokuma et al.(14). All PCR experiments contained one positive control (gDNA of *A. phagocytophilum* infected dog blood) and one negative control (sterilized ddH_2O). The amplification products (5 µl) were electrophoretically resolved in 1.5% agarose gels containing GelRed™ gel stain (Biotium, Inc., Hayward, CA, USA) and were then visualisedon a Fusion FX Gel Documentation System (Vilber Lourmat,

Collégien, France).

All positive products of the Polymerase Chain Reaction (PCR) were purified from an agarose gel for the purpose of sequencing. This was achieved by means of the High Pure PCR product purification kit (Roche, Mannheim, Germany). The purified amplicons were then sequenced in both directions using corresponding PCR primers by Macrogen Inc. (Macrogen, Amsterdam, The Netherlands). The primer sequences were subsequently trimmed from all reads. The forward and reverse sequences were then edited and aligned with Geneious Prime software (http://www.geneious.com) to obtain a single consensus sequence. The final sequences were then aligned to homologous sequences available from GenBank using the BLASTn algorithm with the default settings. The sequence divergences were calculated using a Kimura 2-parameter substitution model15 implemented in the program MEGA X16.

Treatment protocols

Following the diagnosis of pneumonia and anaplasmosis in the herd, oxytetracycline LA (Primamycin LA®, Zoetis) was administered intramuscularly at a dose of 20 mg/kg every 48 hours for a total of three doses. In addition, enrofloxacin (Baytril 10%®, Bayer) was administered subcutaneously at doses of 8 mg/kg, 5 mg/kg, and 5 mg/kg for three consecutive days, respectively, and B complex vitamins (Berovit®, Ceva) were administered intramuscularly at a dose of 5 ml for five consecutive days.

For the purpose of prophylaxis, oxytetracycline LA (Primamycin LA®, Zoetis) was administered intramuscularly to the entire herd at a dose of 20 mg/kg every 48 hours for two doses, followed by a single subcutaneous dose of gamithromycin (Zactran®, Merial) at 6 mg/kg to the whole herd. The decision not to include a negative control group in this study was taken in view of the ethical concerns and the risk of exacerbating morbidity and mortality, as the disease is associated with fatalities.

Statistics

The comparison was made of PCV%, WBC, and WBC counts between the parasitemia-positive and negative groups. This was performed using the SigmaPlot® statistical software. Initially, normality was assessed using the Shapiro-Wilk test, followed by t-tests for group comparisons. A significance level of $P < 0.05$ was considered to be statistically significant in all evaluations.

Results

A physical examination of two goats brought to our hospital, as well as a general examination of the herd, revealed varying degrees of anemia and pneumonia, particularly in both young and adult animals. The primary clinical signs exhibited by the affected animals included weakness, reluctance to move, open-mouth breathing, mucopurulent to purulent nasal discharge, lacrimation, general weakening, painful breathing, and pain during costal and abdominal palpation in some animals. The respiratory rates of the two goats admitted to our hospital were recorded at 136/min and 100/min, respectively, while their heart rates were documented as 148/min and 140/min. Additionally, the body temperatures of the affected animals were measured to be 37.2°C and 37.7°C. It is noteworthy that one of the goats exhibited signs of abdominal discomfort during physical palpation.

In one of the two goats brought to the animal hospital, mild leukocytosis (12,300/mm³) and elevated aspartate transaminase (AST) levels (519 U/L) were observed. In the second goat, significant anemia (PCV 16%) and thrombocytopenia were detected. The second goat exhibited symptoms consistent with severe anemia (PCV 16%), thrombocytopenia (45,000/mm³), neutrophilia (63%), mild leukocytosis (11,200/mm³), and increased levels of AST (338 U/L) and GGT (113 U/L).

A necropsy of deceased goats revealed findings indicative of pneumonia. However, a bacteriological examination of the lungs revealed no identification of *Pasteurella* spp. or *Mycoplasma* spp..

The findings from the two necropsied goats, in conjunction with the clinical and laboratory examinations of other goats in the herd, led to the identification of pneumonia and severe anemia in the affected animals. Parasitological examinations revealed the presence of *A. phagocytophilum* or an *A. capra*-like morula in granulocytes under microscopic analysis, thus necessitating the implementation of molecular diagnostic methods.

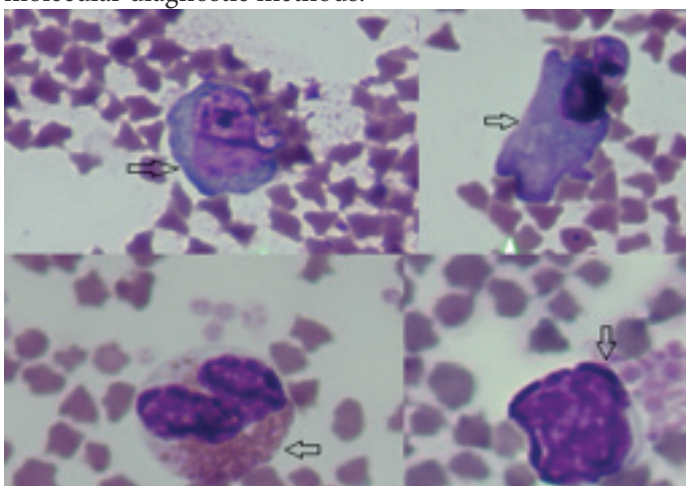

Figure 1. Microscopic view of Anaplasma spp. (Diff-quick stain of blood smear)

The PCV, WBC and PLT levels of a total of 24 goats' blood samples are presented in Table 1. These samples were obtained from two goats that were admitted to the hospital and a subsequent examination of 22 goats in the herd. The samples were examined microscopically for the presence of *Anaplasma* spp., with positive and negative results.

Table 1. Comparison of PCV%, WBC, and PLT levels of microscopically Anaplasma spp. positive and negative goats in the herd.

	***Anaplasma* spp. positive (n=13)**		***Anaplasma* spp.negative (n=11)**		**P value**
	Mean	Min-Max	Mean	Min-Max	
PCV (%)	26,92±1,93	16,00-41,00	28,54±1,13	24,00-36,00	P=0,498
WBC (/mm³)	12.115,0±2.066,0	3.600,0-31.200,0	10.691,0±2.304,0	1.700,0-22.500,0	P=0,649
PLT (/mm³)	155.100,0±58.575,0	39.000,0-660.000,0	265.800,0±81.667,0	54.000,0-468.000,0	P=0,293

All blood samples yielded positive PCR amplifications (Fig. 2). The amplified target fragment of 16S rRNA was successfully sequenced. In the sequence analyses, 305 bp covering the fragments were obtained from all isolates. The BLASTn analysis of the sequences revealed that all the isolates belonged to *Anaplasma* sp.. No intraspecific differences were identified among the *Anaplasma* sp. sequences, which collectively represented a single haplotype based on the 16S rRNA fragment. The sequence of this haplotype has been submitted to GenBank (accession no: MT632469). Furthermore, the partial 16S rRNA sequence of *Anaplasma* sp. isolates exhibited 100% identity with the sequences of previously reported *Anaplasma* isolates from multiple geographical regions.

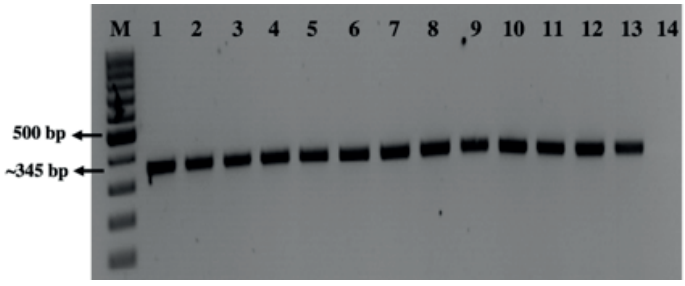

Figure 2. PCR amplification of Anaplasma spp. 16S rDNA partial gene region on gel electrophoresis. M: 100 bp marker; 1-12: positive samples; 13: positive control; 14: negative control.

It is noteworthy that a mere two goats perished as a consequence of their critical condition on the second day of treatment. Subsequent to the conclusion of the treatment regimen, no further fatalities occurred.

Discussion

Anaplasma ovis is the primary species responsible for anaplasmosis in sheep and goats(17,18). Additionally, *A. phagocytophilum*-associated anaplasmosis has been reported in some cases of small ruminants(18,19). Although less common than *A. ovis* and *A. phagocytophilum*, infections with *A. marginale*, the causative agent of cattle anaplasmosis, have been detected in sheep(20) and goats(21). *Anaplasma* capra was first described by Li et al.(5) and has been reported to cause anaplasmosis in sheep and goats(22). In this study, anemia in the herd was suspected to be associated with anaplasmosis based on clinical and laboratory findings. Microscopic examination suggested that the causative agent of anaplasmosis was likely not *A. ovis* but might instead be *A. capra*. Consequently, molecular diagnostic methods were further explored, and the discussion centered on diagnostic techniques and the effectiveness of treatment within the herd.

The diagnosis of anaplasmosis can be made using a variety of methods, including microscopic examination of blood smears; serological methods such as the indirect fluorescent antibody test (IFAT) and competitive ELISA (cELISA); and molecular analyses such as PCR(18,19). However, as asserted by Shabana et al.(19) the reliability of microscopic diagnosis is questionable in non-symptomatic animals and carriers, due to the low number of circulating *Anaplasma* sp.-infected cells. Consequently, the microscopic detection of morulae or agents in peripheral blood is neither sensitive nor reliable for diagnosis(5). It has been stated that IFAT and cELISA techniques are highly effective in diagnosis due to their high specificity and sensitivity, and PCR offers an advantage with its superior sensitivity, enabling the differentiation of *Anaplasma* subtypes(2,6,19,23,24). Peng et al.(25) also stated that the multiplex PCR scanning method can be used for species determination. In accordance with these findings, in our study, only 13 of 24 animals were detected as suspected positive for *Anaplasma* spp. based on blood smear examination, and the definitive presence of the agent was determined only by molecular methods.

In this study, during the microscopic examination, an *A. capra*-like morula was observed, and PCR tests detected the presence of *Anaplasma* species DNA in the samples. However, the BLASTn analysis of partial 16S rRNA sequences was inconclusive in identifying the *Anaplasma* species as *A. capra*, due to the amplification of a conserved gene region. Consequently, the employment of more specific markers, such as the citrate synthase (gltA) or GroEL genes, which exhibit greater sequence variation, is required

to accurately confirm the species(26). The isolate obtained in the present study has been registered in GenBank under the name of Uncultured *Anaplasma* sp. clone AncapTR1 and its accession number is MT632469.

It has been documented that individuals infected with species belonging to the *Anaplasma*taceae family exhibit a range of nonspecific symptoms, including fever, malaise, muscle-joint pain, vomiting, diarrhea, nausea, and anorexia. Laboratory examinations have revealed the presence of leukopenia, thrombocytopenia, elevated lactose dehydrogenase enzyme activities, and increased blood urea nitrogen (BUN) levels(1). Concurrently, Li et al.(5) identified fever, headache, dizziness, chills, and in some cases, muscle pain, rash, nausea, vomiting, diarrhea, regional lymphadenopathy, cough, arthritis, and confusion in a study with *A. capra* infecting 28 people. Laboratory examinations further revealed increased liver enzyme activity (AST and ALT), leukopenia, and thrombocytopenia(5). However, in this research, no complaints about health were reported during the disease period and the following 1 year in the owner and other caregivers of the goat herd. Nevertheless, the increase in liver enzyme activity and signs of leukopenia and thrombocytopenia have been identified in some of the goats which are the subject of our research.

The agents of the genus *Anaplasma* are transmitted transstadially by Ixodid (hard tick) ticks, thus rendering ticks the primary vectors in *Anaplasma* spp. infections(1,5,8,11). Furthermore, studies have demonstrated that migratory birds may play a pivotal role in disseminating *A. phagocytophilum* via *Ixodes ricinus* ticks(27,28). In a study conducted in the Çanakkale region, 14 Ixodid tick species have been identified in sheep, goats, and cattle and their seasonal activities have been demonstrated. The study revealed *Ixodes ricinus* (24.45%) to be most prevalent tick species, followed by *Rhipicephalus bursa* (22.06%), *R. turanicus* (12.61%), *R. (Boophilus) annulatus* (8.8%) and *Dermacentor marginatus* (7.12%), in that order(29). However, given that no ticks were detected on any of the goats in the herd, it could be hypothesized that the transmission occurred through an alternative vector, environmental contamination, or mechanical transmission, a subject that remains open to discussion.

It is generally accepted that the mortality rate of anaplasmosis is not particularly high. However, the present study suggests that the high mortality rate may be attributable to the presence of concomitant pneumonia in the herd or to the occurrence of the disease in Saanen breed goats, resulting in impaired enzootic stability. The treatment of anaplasmosis in sheep and goats has been demonstrated to be effective with drugs such as oxytetracycline, imidocarb dipropionate, and doxycycline(17,18,30). Furthermore, doxycycline is a commonly prescribed treatment for human anaplasmosis(5,7)

A study by Facury-Filho et al.(31) compared the effects of enrofloxacin (7.5 mg/kg single dose; 7.5 mg/kg in two doses, three days apart) and long-acting oxytetracycline (20 mg/kg single dose) in cattle infected with *A. marginale*. The study concluded that both enrofloxacin and oxytetracycline exhibited favorable outcomes in terms of treatment efficacy. However, enrofloxacin demonstrated a more expeditious response in terms of reducing rickettsemia and restoring normal hematocrit levels(31). Furthermore, a separate study by Sing et al.(32) reported that the administration long-acting enrofloxacin (7.5 mg/kg SC, single dose) and oxytetracycline (10 mg/kg, once daily for 5 days IV) resulted in successful treatment outcomes in cattle infected with *A. marginale(*32). Conversely, it has been asserted that enrofloxacin administration at a dose of 12.5 mg/kg SC, administered twice with 48-hour intervals, in splenectomised calves with *A. marginale* infections, demonstrates positive outcomes while failing to eradicate the infection(33). Another study demonstrated that enrofloxacin (5 mg/kg IV q24h for 5 days), imidocarb (5 mg/kg IM twice, 7 days apart), and oxytetracycline (22 mg/kg IV q24h for 5 days) treatments provide some efficacy in clearing persistent *A. marginale* infections in cattle, but none of the treatments eliminate the infection(34). However, no research has been found regarding the use of enrofloxacin in the treatment of anaplasmosis in goats. In the presented study, a combination of oxytetracycline and enrofloxacin was used for the first time to treat *Anaplasma* spp. infections in goats, and the treatment proved to be successful. It is imperative to note that both oxytetracycline and enrofloxacin can be detected in high concentrations in goat milk. Consequently, it is crucial to observe sufficient withdrawal time after their administration to safeguard both the health of the offspring and public health(35,36).

Conclusion

This study confirmed the presence of *Anaplasma* spp. in goats, providing insights into clinical and laboratory findings as well as treatment methods. These results highlight the importance of considering anaplasmosis in cases with similar clinical, microscopic, and molecular characteristics. Additionally, given the zoonotic potential of *Anaplasma* species, precautions should be taken to protect public health, particularly for animal caregivers and veterinarians. The necessity for further research is highlighted by the

paucity of data on the disease in humans, various animal species and breeds, different regions, predisposing factors, and its economic impact. The present study demonstrates the efficacy of a combination of enrofloxacin and oxytetracycline in treating *Anaplasma* infection in goats.

Ethics statements

This research was conducted with the ethical approval of the Committee of Animal Experimentation of Bursa Uludağ University-2020-06/90.

References

1. Fang LQ, Liu K, Li XL, et al. Emerging tick-borne infections in mainland China: an increasing public health threat. Lancet Infect Dis. 2015;15(12):1467-1479. doi:10.1016/S1473-3099(15)00177-2
2. Yang J, Liu Z, Niu Q, et al. A novel zoonotic *Anaplasma* species is prevalent in small ruminants: potential public health implications. Parasit Vectors. 2017;10(1):264. doi:10.1186/s13071-017-2182-9
3. Dantas-Torres F, Chomel BB, Otranto D. Ticks and tick-borne diseases: a One Health perspective. Trends Parasitol. 2012;28(10):437-446. doi:10.1016/j.pt.2012.07.003
4. Seo MG, Kwon OD, Kwag D. Genotypic analysis of piroplasms and associated pathogens from ticks infesting cattle in Korea. Microorganisms. 2020;8(5):728. doi:10.3390/microorganisms8050728
5. Li H, Zheng YC, Ma L, et al. Human infection with a novel tick-borne *Anaplasma* species in China: a surveillance study. Lancet Infect Dis. 2015;15(6):663-670. doi:10.1016/S1473-3099(15)70051-4
6. Peng Y, Wang K, Zhao S, et al. Detection and phylogenetic characterization of *Anaplasma* capra: an emerging pathogen in sheep and goats in China. Front Cell Infect Microbiol. 2018;8:283. doi:10.3389/fcimb.2018.00283
7. Chocklakis D, Ioannou I, Tselentis Y, Psaroulaki A. Human anaplasmosis and *Anaplasma* ovis variant. Emerg Infect Dis. 2010;16(6):1031-1032. doi:10.3201/eid1606.090175
8. Rar V, Golovljova I. *Anaplasma*, Ehrlichia, and "Candidatus Neoehrlichia" bacteria: pathogenicity, biodiversity, and molecular genetic characteristics, a review. Infect Genet Evol. 2011;11(8):1842-1861. doi:10.1016/j.meegid.2011.09.019
9. Maggi RG, Mascarelli PE, Havenga LN, Naidoo V, Breitschwerdt EB. Co-infection with *Anaplasma* platys, Bartonella henselae and Candidatus *Mycoplasma* haematoparvum in a veterinarian. Parasit Vectors. 2013;6:103. doi:10.1186/1756-3305-6-103
10. Kadkhoda K, Gretchen A. Retrospective study investigating the seroprevalence of *Anaplasma* phagocytophilum in Manitoba, Canada: 2011–2014. Open Forum Infect Dis. 2016;3(4):ofw199. doi:10.1093/ofid/ofw199
11. Battilani M, De Arcangeli S, Balboni A, Dondi F. Genetic diversity and molecular epidemiology of *Anaplasma*. Infect Genet Evol. 2017;49:195-211. doi:10.1016/j.meegid.2017.01.021
12. Amer S, Kim S, Yun Y, Na KJ. Novel variants of the newly emerged *Anaplasma* capra from Korean water deer (Hydropotes inermis argyropus) in South Korea. Parasit Vectors. 2019;12(1):365. doi:10.1186/s13071-019-3622-5
13. Parola P, Roux V, Camicas JL, et al. Detection of ehrlichiae in African ticks by polymerase chain reaction. Trans R Soc Trop Med Hyg. 2000;94(6):707-708. doi:10.1016/S0035-9203(00)90243-8
14. Inokuma H, Raoult D, Brouqui P. Detection of Ehrlichia platys DNA in brown dog ticks (Rhipicephalus sanguineus) in Okinawa Island, Japan. J Clin Microbiol. 2000;38(11):4219-4221.
15. Kimura M. A simple method for estimating evolutionary rate of base substitutions through comparative studies of nucleotide sequences. J Mol Evol. 1980;16:111-120.
16. Kumar S, Stecher G, Li M, Knyaz C, Tamura K. MEGA X: Molecular evolutionary genetics analysis across computing platforms. Mol Biol Evol. 2018;35(6):1547-1549. doi:10.1093/molbev/msy096
17. Smith MC, Sherman DM. Goat Medicine. 2nd ed. Wiley-Blackwell; 2009:287-288.
18. Karatepe M, Karatepe B. Anaplasmosis. In: Özcel MA, ed. Veteriner Hekimliğinde Parazit Hastalıkları. Cilt 2. Türkiye; 2013:812-815.
19. Shabana II, Alhadlag NM, Zaraket H. Diagnostic tools of caprine and ovine anaplasmosis: a direct comparative study. BMC Vet Res. 2018;14(1):165. doi:10.1186/s12917-018-1489-x
20. Yousefi A, Rahbari S, Shayan P, Sadeghi-dehkordi Z, Bahonar A. Molecular detection of *Anaplasma* marginale and *Anaplasma* ovis in sheep and goats in west highland pasture of Iran. Asian Pac J Trop Biomed. 2017;7(5):455-459. doi:10.1016/j.apjtb.2017.01.017
21. Da Silva NB, Taus NS, Johnson WC, et al. First report of *Anaplasma* marginale infection in goats, Brazil. PLoS One. 2018;13(8):e0202140. doi:10.1371/journal.pone.0202140
22. Yang J, Han R, Niu Q, et al. Occurrence of four *Anaplasma* species with veterinary and public health significance in sheep, northwestern China. Ticks Tick-Borne

Dis. 2018;9(1):82-85. doi:10.1016/j.ttbdis.2017.10.005
23. Seo HJ, Jin BC, Kim KH, et al. Molecular Detection and Phylogenetic Analysis of *Anaplasma* spp. in Korean Native Goats from Ulsan Metropolitan City, Korea. Vector-Borne Zoonotic Dis. 2019;19(10):773-776. doi:10.1089/vbz.2018.2374
24. Jouglin M, Blanc B, de la Cotte N, et al. First detection and molecular identification of the zoonotic *Anaplasma* capra in deer in France. PLoS One. 2019;14(7):e0219184. doi:10.1371/journal.pone.0219184
25. Peng Y, Zhao S, Wang K, et al. A Multiplex PCR Detection Assay for the Identification of Clinically Relevant *Anaplasma* Species in Field Blood Samples. Front Microbiol. 2020;11:606. doi:10.3389/fmicb.2020.00606
26. Lin ZT, Ye RZ, Liu JY, et al. Epidemiological and phylogenetic characteristics of emerging *Anaplasma* capra: A systematic review with modeling analysis. Infect Genet Evol. 2023;115:105510. doi:10.1016/j.meegid.2023.105510
27. Alekseev AN, Dubinina HV, Semenov AV, Bolshakov CV. Evidence of ehrlichiosis agents found in ticks (Acari: Ixodidae) collected from migratory birds. J Med Entomol. 2001;38(4):471-474. doi:10.1603/0022-2585-38.4.471
28. Stuen S, Granquist EG, Silaghi C. *Anaplasma* phagocytophilum–a widespread multi-host pathogen with highly adaptive strategies. Front Cell Infect Microbiol. 2013;3:31. doi:10.3389/fcimb.2013.00031
29. Aydın L. Güney Marmara Bölgesi ruminantlarında görülen kene türleri ve yayılışları. T Parazitol Derg. 2000;24(1):194-200.
30. Stuen S, Longbottom D. Treatment and Control of Chlamydial and Rickettsial Infections in Sheep and Goats. Vet Clin N Am-Food Anim. 2011;27(1):213-233. doi:10.1016/j.cvfa.2010.10.017
31. Facury-Filho EJ, de Carvalho AÚ, Ferreira PM, et al. Effectiveness of enrofloxacin for the treatment of experimentally-induced bovine anaplasmosis. Rev Bras Parasitol Vet. 2012;21(1):32-36. doi:10.1590/s1984-29612012000100007
32. Singh ST, Randhawa SS, Kaur S, et al. Therapeutic efficacy of long acting enrofloxacin in anaplasmosis in cattle. Indian Vet J. 2015;92(3):80-81.
33. Coetzee JF, Apley MD. Efficacy of enrofloxacin against severe experimental *Anaplasma* marginale infections in splenectomized calves. Vet Ther. 2006;7(3):319-328.
34. Coetzee JF, Apley MD, Kocan KM. Comparison of the efficacy of enrofloxacin, imidocarb, and oxytetracycline for clearance of persistent *Anaplasma* marginale infections in cattle. Vet Ther. 2006;7(4):347-360.
35. Zakaria AM, Al-Daek T, Elmeligy E, et al. Effect of different post-partum therapeutic protocols with intrauterine oxytetracycline, oxytocin and/or GnRH injection in post-kidding goats on oxytetracyclines residues in goat milk and postpartum ovarian resumption with referring to clinical and haematological pictures. BMC Vet Res. 2023;19:139. doi:10.1186/s12917-023-03706-0
36. Quintanilla P, Beltrán MC, Molina MP, Escriche I. Enrofloxacin treatment on dairy goats: Presence of antibiotic in milk and impact of residue on technological process and characteristics of mature cheese. Food Control. 2021;112:107762. doi:10.1016/j.foodcont.2020.107762