CHAPTER 07

ANAPLASMOSIS

Saba Mehnaz*[1], Rao Zahid Abbas[1], Farhan Ahmad Atif[2], Tabassam Fatima[3], Muhammad Arfan Zaman[4], Nauman Rafique[1] and Umer Farooq[4]

[1]Department of Parasitology, University of Agriculture, Faisalabad.
[2]Medicine Section, Department of Clinical Sciences, College of Veterinary and Animal Sciences, Jhang, Sub-Campus University of Veterinary and Animal Sciences, Lahore 54600, Pakistan.
[3]Department of Parasitology, Riphah College of Veterinary Sciences, Riphah International University, Lahore.
[4]Department of Pathobiology, College of Veterinary and Animal Sciences, Jhang, Sub-Campus University of Veterinary and Animal Sciences, Lahore 54600, Pakistan.
*Corresponding author: sabamehnaz2012@gmail.com

INTRODUCTION

Anaplasmosis is caused by obligatory intra-erythrocytic bacteria called Anaplasma (Order: Rickettsiales, Family: Anaplasmatacae, Genus: Anaplasma). The disease caused by Anaplasma is called anaplasmosis and is characterized by anemia, high fever, jaundice, abortion and death (Waruri et al. 2021). There are different species of genus Anaplasma (A.) cause disease in humans and animals including *A. marginale*, *A. phago¬cytophilum* and *A. ovis* (Fishbein et al. 1994; Dumler and Bakken 1998; Dumler and Broqui 2004; Zobba et al. 2020). Anaplasmosis is transmitted through infected tick-bite, biting flies and contaminated fomites. Bovine anaplasmosis is commonly known as gall sickness and is prevalent throughout the world (Rochlin and Toledo 2020).

Etiology

The word Anaplasma is a combination of Greek words ana means without and plasma means molded or produce (Dumler et al. 2004; Atif et al. 2021). On the basis of phylogenetic analysis, family Anaplasmataceae includes following genera Anaplasma, Aegyptianella, Candidatus, Neorickettsia, Ehrlichia and Wolbachieae (NCBI gene bank information). Genus Anaplasama comprises of various species including A. marginale, *A. centrale*, *A. ovis*, *A. bovis*, *A. phagocytophilum*, *A. platys* and *A. odocoilei* (Dumler et al. 2001). *A. marginale*, *A. centrale*, *A. bovis* and A. ovis are the species prevalent in ruminants (Table 1). *A. phagocytophilum* causes granulocytic anaplasmosis in both equines (EGA-Equine Granulocytic Anaplasmosis) and humans (HGA-Human Granulocytic Anaplasmosis).
Development of Anaplasma spp. occurs in the vertebrate host and arthropod vector. In case of vertebrate host, Anaplasma persists in their bodies for long duration and they act as reservoir host. When a vector feed on infected reservoir host, becomes infected. Anaplasma spp. grows in gut cells as well as in salivary glands of ticks and is transmitted through tick saliva during blood feeding (Villar et al. 2016).
Occurrence of species of genus Anaplasma is sporadic throughout the world but endemic in tropical and subtropical regions of world due to high tick burden in these areas and high risk of carrier animals (Smith et al. 1986; Wickwire et al. 1987; Allred et al. 1990; Rodriguez et al. 2000; de la Fuente et al. 2001; Palmer 2001; Maggi and Krämer 2019). *A. marginale* infected final hosts as well as vectors (ticks) acts as reservoirs of infection to other susceptible hosts. *A. marginale* only resides inside the cytoplasmic vacuoles of erythrocytes (Richey 1981; Raboloko et al. 2020). About four rickettsias are present in each erythrocyte while more than 70% of erythrocytes become infected with Anaplasma during the acute phase of the disease. Reticuloendothelial system removes infected erythrocytes which results in development of anemia and icterus (Richey 1981; Saetiew et al. 2020).

Transmission of Anaplasma Species

The mode of Anaplasma species transmission is based on the ecological conditions and availability of final host. Different geographical strains of Anaplasma are identified which have different morphological features, characteristics of antigen and their ability to disseminate by vectors (Cabezas-Cruz et al. 2013).
Various routes observed for the transmission of Anaplasma species includes vectors, fomites and transplacental transmission. Ticks play the role of biological vector whereas, flies are mechanical vectors. There are about twenty species of ticks; belonging to genus Rhipicephalus and Dermacentor, involved in the transmission of Anaplasma species (Aubry and Geale 2011). Unlike Genus Babesia, trans-ovarian transmission (transmission from female ticks to larvae) of Anaplasma species does not occur in ticks. So, persistent infection in host is needed for spread of Anaplasma species. Hematophagous flies including families Culicidae, Muscidae and Tabanidae are important in the transmission process of Anaplasma species. Fomites include surgical instrument, needles and piercing objects. Infected mothers may pass the infection to their claves, which end up in calf mortality (Kocan et al. 2010). It is noteworthy that there is still a conflict on transplacental transmission of Anaplasma species as some reports did not verify transplacental transmission (Aubry and Geale 2001). This conflict could be due to genetic variations among Anaplasma species and level of host immunity.

How to cite this chapter: Mehnaz S, Abbas RZ, Atif FA, Fatima T, Zaman MA, Rafique N and Farooq U, 2022. Anaplasmosis. In: Abbas RZ, Khan A, Liu P and Saleemi MK (eds), Animal Health Perspectives, Unique Scientific Publishers, Faisalabad, Pakistan, Vol. I, pp: 47-53. https://doi.org/10.47278/book.ahp/2022.07

Table 1: Molecular diagnosis of various species of genus *Anaplasma*

Species	Molecular test	Animal screened	Reference
A. phagocytophilum	Nested PCR	Goat	Tumwebaze et al. 2020
A. ovis	PCR, PCR-RFLP	Goat	Aguirre et al. 2006
A. marginale	PCR-RFLP	Cattle	Ayyez et al. 2019
Anaplasma spp.	PCR, ELISA, Nested PCR	Sheep	Fuente et al. 2001

Characterization of Genus Anaplasma

Below given is the detailed description of various species of genus Anaplasma:

A. marginale

A. marginale is an obligate species of Anaplasma which causes 'bovine anaplasmosis' or 'gall sickness'. A variety of wild and domestic ruminants are infected by A. marginale including Bos taurus, Bos indicus, Bubalus bubalis, Cervids and Giraffa etc. (Kocan 2003; Aubry and Geale 2011; Ayyez et al. 2019). Unlike transovarial transmission of Babesia, transstadial transmission occurs in case of *A. marginale* (Kocan et al. 1981; Stich et al. 1989; Kocan et al. 1992; Madison-Antenucci 2020). Transmission of *A. marginale* occurs through ticks, biting flies, fomites. Moreover, tans-placental transmission occurs during second and third trimester of pregnancy (Dikmans 1950; Zaugg 1985; Atif et al. 2021). More than 20 species of ticks are involved in transmission of *A. marginale* (Dikmans 1950; Kocan 2003; Da Silva et al. 2015). The mechanical routes of *A. marginale* transmission are significant in tick free areas (Dikmans 1950; Da Silva et al. 2013; Solomon and Tanga 2020). Clinical signs of gall sickness include high fever, emaciation, abortion, lethargy and death in few cases (Barbet et al. 1999; Kocan 2003; Sazmand et al. 2020). Incubation period of *A. marginale* is variable from days to months. During acute phase of infection, *A. marginale* infects up to 70% erythrocytes. During blood smear examination of infected animals, 2 to 4 intra-erythrocytic inclusion bodies are seen, and this could be major reason of high erythrocytic destruction (Barbet et al.1999; Kocan et al. 2010; Berthelsson et al. 2020).

A. centrale

A. central, is morphologically similar to *A. marginale* but it resides inside the erythrocytes but its inclusions are more central (Khodadadi et al. 2021). *A. centrale* was identified by Sir Arnold Theiler in 1911. Prevalence of A. centrale is high in tropical and subtropical areas (Rar and Golovljova 2011; Sarangi et al. 2021). It causes mild anemia in infected animals being less pathogenic by its nature and that is why it is preferred to be used as vaccine agent against anaplasmosis (Zaugg 1985; Kocan 2003; Rajput et al. 2005; Abdullah et al. 2021). *A. centrale* transmission occurs through ticks, biting flies and fomites. Only tick species involved in transmission of *A. centrale* is Rhipicephalus simus (Calleja-Bueno 2021) but with the help of molecular techniques, its presence in Haemaphysalis punctate and Amblyomma sp., has also been reported (Palomar et al. 2015; Teshale et al. 2015). *A. centrale* cause mild disease in cattle (Rar and Golovljova 2011). For control of bovine anaplasmosis live attenuated vaccine of *A. centrale* is being used in Australia, Africa, Latin America and Israel (Kocan et al. 2010; Waruri et al. 2021).

A. bovis

A. bovis was first time identified in cattle in 1936. Unlike *A. marinale* and *A. centrale*, *A. bovis* is also an obligate intra-monocytic species of Anaplasma in cattle (Donatien and Lestoquard 1936). Prevalence of *A. bovis* has been reported in United States, Europe, Asia and Africa (Uilenberg 1995; Goethert and Telford 2003; Kawahara et al. 2006; Santos and Carvalho 2006; Ceci et al. 2014; García-Pérez 2016; Consolaro et al. 2019). *A. bovis* can cause disease in cattle, buffalo, goat, dog, deer, Mongolian gazelle, raccoon, leopard cat, cotton-tail rabbit and eastern rock sengi but bovines (cattle and buffalo) are the usual host (Goethert and Telford 2003; Guo et al. 2020). Tick genera involved in its transmission are Amblyomma, Hyalomma, Rhipicephalus and Haemaphysalis (Uilenberg 1995; Dumler et al. 2001; Goethert and Telford 2003; Harrus and Waner 2011; Palomar et al. 2015). Clinical signs develop in infected cattle and buffalo include fever, emaciation, decrease in weight gain, icteric mucous membranes, inflamed pre-scapular lymph node and sometime death may occur in later stages of the disease (Donatien and Lestoquard 1936; Uilenberg 1995; Uilenberg 1997; Santos and Carvalho 2006; Arulkumar et al. 2016).

A. ovis

A. ovis is an obligate intra-erythrocytic species of Anaplasmain sheep. It was first identified by Bevan in 1912 (Bevan 1912; Dumler et al. 2001; Peng et al. 2021). *A. ovis* is prevalent in USA, Asia, Africa and Europe (Al-Hosary et al. 2021). *A. ovis* cause disease in small ruminants including wild ruminants (Bevan 1912; Kuttler 1984; Friedhoff 1997; de la Fuente et al. 2008; Li et al. 2015; Enkhtaivan et al. 2019). It also has zoonotic importance as it has been detected in humans by Chochlakis et al. (2010). Intra-erythrocytic inclusions observed during A. ovis, are observed at central position in 35–40% cases and at marginal positions of erythrocytes in 60 to 65% cases (Shompole et al. 1989; Primo et al. 2019). Clinical signs observed during the course of the disease are emaciation, anemia, fever, depression, rumen atony, decreased production, abortion and death of the animal (Yousif et al. 1983; Manickam 1987; Friedhoff 1997; Yasini et al. 2012; Tumwebaze et al. 2020). These signs of the disease depend on the breed, age and body condition score of the animal (Splitter et al. 1955; Shompole et al. 1989; Tumwebaze et al. 2020). Development of anemia due to A. ovis is the main cause of huge economic losses at livestock farms especially in developing countries, as sheep and goats are mainly reared in areas of tropics and subtropics (Jensen 1955; Zaugg 1985; Lacasta et al. 2021).

A. platys

A. platys, an also an obligate intracellular pathogen which resides in platelets, was observed initially during blood examination of dog in U.S.A. (Florida) in 1978. Later on, it was detected in almost all continents (Harvey et al. 1978; Suksawat

et al. 2001; Sanogo et al. 2003; Sparagano et al. 2003; Brown 2008; Aguirre et al. 2006; de la Fuente et al. 2006; Kawahara et al. 2006; Melo et al. 2016). A. platys mainly affect canines but it has been also reported in foxes, cats, camels, cattle, deer and humans (Harvey et al. 1978; Maggi et al. 2013; Qurollo et al. 2014; Cardoso et al. 2015; Dahmani et al. 2015; Li et al. 2015; André et al. 2020). In canines, A. platys causes the disease namely Canine Cyclic Thrombocytopenia (CCT). Disease is characterized by fever, severe emaciation, loss of appetite, lethargy, respiratory distress, increased mucus secre¬tion and ocular discharge, muzzle hyperkeratosis and splenomegaly (Sainz et al. 2015; Brandão 2019). Severity and type of clinical signs depend mainly on dog species. Transmission of A. platys occurs through tick species: Rhipicephalus sanguineus, Rhipicephalus turanicus, Dermacentor auratus, Haemaphysalis longicornis, Ixodes persulcatus, Haemaphysalis longicornis, chewing louse, Heterodoxus spiniger and blood transfusion (Harvey et al. 1978; Parola et al. 2003; Martin et al. 2005). Severe thrombocytopenia is observed during the course of disease in dogs which may reoccur after two weeks of incomplete recovery. Thrombocytopenia probably occurs due to direct damage of platelets by pathogen and immune mediated mechanism (immune cells) (Harvey et al. 1978; Piratae et al. 2019). Clinical signs of the disease are fever, anorexia, emaciation, pale mucus membranes, lethargy, pin point hemorrhages on oral mucosa and skin, lymph adenomegaly and epistaxis (Bradfield et al. 1996; Bouzouraa et al. 2016).

A. phagocytophilum

A. phagocytophilum, found in granulocytes of host, has great importance with respect to veterinary and public health (Dugat et al. 2015). A sheep was affected with a tick-borne disease of unknown etiological agent during 1932 in Scotland whose causative agent was described in 1940 as *A. phagocytophilum*. The host range of *A. phagocytophilum* includes human, carnivores, ruminants, reptiles, birds and rodents. Ehrlichia was known to cause disease in humans and horses and previously it was known as Ehrlichia equi (Stuen et al. 2002). During the course of the disease about 40% of the patients need hospitalization. Rate of the mortality of disease is 7-10 % in USA but in Europe there is no mortality (Fishbein et al. 1994; Dumler and Bakken 1998; Blanco and Oteo 2002; Dumler and Broqui 2004). Situation aggravates leading to death in case of prevailing health issues viz; intravascular coagulation, renal failure, cardiac enlargement, coma and seizures. The disease is more harmful in immune-compromised patients of old age and children (Fishbein et al. 1994; Dumler and Bakken 1998; Rocco et al. 2020). In ruminants, disease is characterized by rise in body temperature, in appetence, drop in milk and meat production and abortion. Death occurs in infected animals which remains unattended during the course of the disease (Stuen et al. 2002; Rodino et al. 2020). *A. phagocytophilum* is transmitted by ticks belonging to genera Ixodes, Dermacentor, Haemaphysalis and Amblyomma in USA, Europe and Asia (Holden 2003; Santos et al. 2004; Woldehiwet 2010; Clark 2012; Paulauskas et al. 2012; Tomanović et al. 2013; Rochlin and Toledo 2020). Transstidial transmission of *A. phagocytophilum* has also been reported (Baldridge et al. 2009; Dugat et al. 2015). *A. phagocytophilum* leads to granulocytic anaplasmosis in ruminants and clinical signs include fever, anorexia, abortion and decrease in milk production (Woldehiwet 2006; Stuen et al. 2002; Dugat et al. 2015). Furthermore, it leads to decrease in immune status of the animal which tends to increase in susceptibility of animal towards secondary or opportunistic bacterial infection (Woldehiwet 2006; Kahn et al. 2019).

Pathogenesis

Anaplasmosis is commonly categorized into four phases; incubation phase, developmental phase, convalescent and carrier phase. Incubation phase is the duration from inoculation of pathogen to a susceptible host until 1% of erythrocytes become infected (clinical signs develop). The duration of each phase of the disease is directly linked with the total number of pathogens entered into host animal. During this stage the PCV remains constant; erythrocytes are produced at the same rate as they are destroyed and no clinical signs are seen. The developmental stage is characterized by onset of clinical signs associated with mortality and sporadic abortions (Coetzee et al. 2010). Clinically recovered animals develop persistent infections and remain as carriers with undetectable parasitemia consequently act as reservoir of infectious agent (Kieser et al. 1990). The pathogen enters erythrocytes through endocytosis and exit by exocytosis and attack on other fresh erythrocytes. Infected erythrocytes are destroyed by reticulo-endothelial cells which leads to development of hemolytic anemia and icterus. Oxidative stress occurs due to imbalance in antioxidants and oxidant ratio. Due to increased production of reactive oxygen species (ROS), there is increase in oxidant level on cellular level. This increased level of oxidants leads to cellular damage and lipid peroxidation "LPO".

Economic Importance of Anaplasma

Anaplasma can cause huge economic losses in terms of decrease in productivity of animal, abortion, increase in treatment cost and mortality of infected animal. It can also cause death in new borne calves because of its trans-placental transmission. According to FAO report, the rate of mortality in case of anaplasmosis is 5 to 70%. In Norway, *A. marginale* leads to mortality of more than 0.3 million lambs each year (Stuen et al. 2018).

Diagnosis

Parasitological diagnosis provides direct evidence of Anaplasma infection. It includes microscopic examination of ethanol fixed Giemsa-stained thin blood smears. This is regarded as the best method of identifying *A. marginale* and to determine the parasitemias of the animals (Yoshihara et al. 2003; Siddiki et al. 2010). Inoculation of susceptible laboratory animals with blood of suspected animals to demonstrate presence of *A. marginale* is another technique (Siddiki et al. 2010). Nucleic acid detection methods for Anaplasma infection includes PCR, qPCR, RT PCR and LAMP. 16S ribosomal gene is the most targeted gene for the detection of anaplasma species (Atif et al. 2021). Whole blood preserved in EDTA coated tubes can be used for the diagnosis of anaplasma species (Hebels et al. 2014). Now a days rapid and sensitive real time assay is using more as compared to nested PCR (Atif et al. 2021).

Treatment

Doxycycline is the drug of choice for treatment of *A. platys* infection in canine (Sainz et al. 2015; Paterson et al. 2020). For

control of bovine anaplasmosis live attenuated vaccine of A. centrale is being used in Australia, Africa, Latin America and Israel (Kocan et al. 2010; Waruri et al. 2021).

Conclusion

Although Anaplasmosis has history of more than a century, still it is one of the most challenging parasites of human and animal health due to variety of species that are present in different hosts. Anaplasmosis is a bacterial disease that causes severe infection and leads to liver damage, severe anemia occurs due to lysis of erythrocytes which causes death of the animals. Disease is more prevalent in young animals. The only way to combat anaplasmosis is to adopt good precautionary measures.

REFERENCES

Abdullah HHAM et al., 2021. Multiple vector-borne pathogens of domestic animals in Egypt. PLoS Neglected Tropical Diseases15: Article # 0009767.

Aguirre E et al., 2006. Genetic characterization of Anaplasma (Ehrlichia) platys in dogs in Spain. Journal of Veterinary Medicine, Series B 53: 197-200.

Al-Hosary AA et al., 2021. Phylogenetic study of *Theileria ovis* and *Theileria lestoquardi* in sheep from Egypt: Molecular evidence and genetic characterization. Veterinary World 14: 634-639.

Allred DR et al., 1990. Molecular basis for surface antigen size polymorphisms and conservation of a neutralization-sensitive epitope in *Anaplasma marginale*. Proceedings of the National Academy of Sciences 87: 3220-3224.

André MR et al., 2020. The co-infection with *Ehrlichia minasensis, Anaplasma marginale* and *Anaplasma platys* is not associated with anemia in beef cattle in the Brazilian Pantanal. Veterinary Parasitology: Regional Studies and Reports 21: 100437.

Arulkumar T et al., 2016. Alpha proteobacteria of genus Anaplasma (Rickettsiales: Anaplasmataceae): Epidemiology and characteristics of Anaplasma species related to veterinary and public health importance. Parasitology 143: 659-685.

Atif FA et al., 2021. Strategies for prevention and control of anaplasmosis: at human-animal interface. Pakistan Journal of Agricultural Science 58: 1649-1662.

Aubry P and Geale DW, 2011. A review of bovine anaplasmosis. Transboundary and Emerging Diseases 58: 1-30.

Ayyez HN et al., 2019. Molecular detection and phylogenetic analysis of Anaplasma Spp. in cattle in Al-Qadisiyah province of Iraq. Macedonian Veterinary Review 42: 181-188.

Baldridge GD et al., 2009. Transovarial transmission of Francisella-like endosymbionts and Anaplasma phagocytophilum variants in Dermacentor albipictus (Acari: Ixodidae). Journal of Medical Entomology 46: 625-632.

Barbet AF et al., 1999. Comparison of surface proteins of Anaplasma marginale grown in tick cell culture, tick salivary glands, and cattle. Infection and Immunity 67: 102-107.

Berthelsson J et al., 2020. *Anaplasma ovis* infection in goat flocks around Gaborone, Botswana. Comparative Clinical Pathology 29: 167-172.

Bevan LLEW, 1912. Anaplasmosis of sheep. The Veterinary Journal 68: 400-401.

Blanco JR and Oteo JA, 2002. Human granulocytic ehrlichiosis in Europe. Clinical Microbiology and Infection 8: 763-772.

Bouzouraa T et al., 2016. Clinical and laboratory features of canine Anaplasma platys infection in 32 naturally infected dogs in the Mediterranean basin. Ticks and Tick-Borne Diseases 7: 1256-1264.

Bradfield JF et al., 1996. *Ehrlichia platys* infection on dogs. Journal of Laboratory Animal Science 46: 565-568.

Brandão VMD, 2019. Molecular detection of *Ehrlichia canis* and *Anaplasma platys* in dogs from municipality of Belém, State of Pará, Brazil. Ciência Rural 49: Article # 20190414.

Brown WC, 2008. Unraveling the immune regulatory mechanisms imposed by Anaplasma. Veterinary Journal 175: 10-11.

Cabezas-Cruz A et al., 2013. Functional and immunological relevance of *Anaplasma marginale* major surface protein 1a sequence and structural analysis. PloS ONE, 8: e65243.

Calleja-Bueno L, 2021. First detection of *Anaplasma phagocytophilum* and *Babesia divergens* and high infection rates of *A. marginale* and *B. bigemina* in cattle in extensive grazing systems of Central Spain. Transboundary and Emerging Diseases 10: Article # 14

Cardoso L et al., 2015. First report of *Anaplasma platys* infection in red foxes (Vulpes vulpes) and molecular detection of *Ehrlichia canis* and Leishmania infantum in foxes from Portugal. Parasites and Vectors 8: 1-8.

Ceci L et al., 2014. Retrospective study of haemoparasites in cattle in Southern Italy by reverse line blot hybridization. Journal of Veterinary Medical Science 76: 869-875.

Chochlakis D et al., 2010. Human anaplasmosis and *Anaplasma ovis* variant. Emerging Infectious Diseases 16: 1031-1032.

Clark KL, 2012. *Anaplasma phagocytophilum* in small mammals and ticks in northeast Florida. Journal of Vector Ecology 37: 262-268.

Coetzee JF et al., 2010. Seroprevalence of *Anaplasma marginale* in 2 Iowa feedlots and its association with morbidity, mortality, production parameters, and carcass traits. The Canadian Veterinary Journal 51: 862-868.

Consolaro A et al., 2019. Phenotypic variability and disparities in treatment and outcomes of childhood arthritis throughout the world: an observational cohort study. The Lancet Child and Adolescent Health 3: 255-263.

Da Silva et al., 2013. Lice outbreak in buffaloes: evidence of *Anaplasma marginale* transmission by sucking lice Haematopinus tuberculatus. The Journal of Parasitology 99: 546-547.

Da Silva JB et al., 2015. Molecular characterization of *Anaplasma marginale* in ticks naturally feeding on buffaloes. Infection, Genetics and Evolution 35: 38-41.

Dahmani M et al., 2015. Development of a new PCR-based assay to detect Anaplasmataceae and the first report of *Anaplasma phagocytophilum* and *Anaplasma platys* in cattle from Algeria. Comparative Immunology, Microbiology and Infectious Diseases 39: 39-45.

de la Fuente J et al., 2006. Reduction of tick infections with *Anaplasma marginale* and *A. phagocytophilum* by targeting the tick protective antigen subolesin. Parasitology Research 100: 85-91.

de la Fuente J et al., 2008. Overview: ticks as vectors of pathogens that cause disease in humans and animals. Frontiers in Bioscience 13: 6938-6946.

de la Fuente J et al., 2001. Molecular phylogeny and biogeography of North American isolates of *Anaplasma marginale* (Rickettsiaceae: Ehrlichieae). Veterinary Parasitology 97: 65-76.

Dikmans G, 1950. The transmission of anaplasmosis. American Journal of Veterinary Research 11: 5-16.

Donatien LF, 1936. Rickettsia bovis, nouvelle espece pathogene pour le boeuf. Bulletin de la Societe de Pathologie Exotique 29: 1057–1061.

Dugat T et al., 2015. Opening the black box of *Anaplasma phagocytophilum* diversity: current situation and future perspectives. Frontiers in Cellular and Infection Microbiology 5: 61-73.

Dumler JS and Bakken JS, 1998. Human ehrlichioses: newly recognized infections transmitted by ticks. Annual Review of Medicine 49: 201-213.

Dumler JS et al., 2001. Reorganization of genera in the families Rickettsiaceae and Anaplasmataceae in the order Rickettsiales: unification of some species of Ehrlichia with Anaplasma, Cowdria with Ehrlichia and Ehrlichia with Neorickettsia, descriptions of six new species combinations and designation of *Ehrlichia equi* and 'HGE agent' as subjective synonyms of *Ehrlichia phagocytophila*. International Journal of Systematic and Evolutionary Microbiology 51: 2145-2165.

Dumler JS and Broqui P, 2004. Molecular diagnosis of human granulocytic anaplasmosis. Expert Review of Molecular Diagnostics 4: 89–98.

Enkhtaivan B et al., 2019. Molecular detection of *Anaplasma ovis* in small ruminants and ixodid ticks from Mongolia. Parasitology International 69: 47-53.

Fishbein DB et al., 1994. Human ehrlichiosis in the United States, 1985 to 1990. Annals of Internal Medicine 120: 736-743.

Friedhoff KT, 1997. Tick-borne diseases of sheep and goats caused by Babesia, Theileria or Anaplasma spp. Parassitologia 39: 99-109.

García-Pérez AL, 2016. Anaplasmataceae in wild ungulates and carnivores in northern Spain. Ticks and Tick-Borne Diseases 7: 264-269.

Goethert HK and Telford III SR, 2003. Enzootic transmission of *Anaplasma bovis* in Nantucket cotton tail rabbits. Journal of Clinical Microbiology 41: 3744-3747.

Guo WP et al., 2020. Extensive genetic diversity of *Anaplasma bovis* in ruminants in Xi'an, China. Ticks and Tick-Borne Diseases 11: Article # 101477.

Harrus S and Waner T, 2011. Diagnosis of canine monocytotropic ehrlichiosis (Ehrlichia canis): an overview. The Veterinary Journal 187: 292-296.

Harvey et al., 1978. Cyclic thrombocytopenia induced by a Rickettsia-like agent in dogs. Journal of Infectious Diseases 137: 182-188.

Holden K, 2003. Detection of Borrelia burgdorferi, *Ehrlichia chaffeensis*, and *Anaplasma phagocytophilum* in ticks (Acari: Ixodidae) from a coastal region of California. Journal of Medical Entomology 40: 534-539.

Hebels DG et al., 2014. Elimination of heparin interference during microarray processing of fresh and biobank-archived blood samples. Environmental and Molecular Mutagenesis, 55: 482-491.

Jensen R, 1955. Suspected anaplasmosis in sheep. North American Veterinarian 36: 117-118.

Kahn LH et al., 2019. From farm management to bacteriophage therapy: strategies to reduce antibiotic use in animal agriculture. Annals of the New York Academy of Sciences 1441: 31-39.

Kawahara M et al., 2006. Novel genetic variants of *Anaplasma phagocytophilum, Anaplasma bovis, Anaplasma centrale*, and a novel Ehrlichia sp. in wild deer and ticks on two major islands in Japan. Applied and Environmental Microbiology 72: 1102-1109.

Khodadadi N et al., 2021. Identification of *Anaplasma marginale* in long-eared hedgehogs (Hemiechinus auritus) and their Rhipicephalus turanicus ticks in Iran. Ticks and Tick-Borne Diseases 12: Article # 101641.

Kieser ST et al., 1990. Cyclic rickettsemia during persistent *Anaplasma marginale* infection in cattle. Infectious Immunology 58: 1117–1119.

Kocan KM et al., 1981. Transmission of *Anaplasma marginale* Theiler by Dermacentor andersoni Stiles and Dermacentor variabilis (Say). American Journal of Veterinary Research 42: 15-18.

Kocan KM et al., 1992. Persistence of *Anaplasma marginale* (Rickettsiales: Anaplasmataceae) in male Dermacentor andersoni (Acari: Ixodidae) transferred successively from infected to susceptible calves. Journal of Medical Entomology 29: 657-668.

Kocan KM, 2003. Antigens and alternatives for control of *Anaplasma marginale* infection in cattle. Clinical Microbiology Reviews 16: 698-712.

Kocan KM et al., 2010. The natural history of *Anaplasma marginale*. Veterinary Parasitology 167: 95-107.

Kuttler KL, 1984. Anaplasma infections in wild and domestic ruminants: a review. Journal of Wildlife Diseases 20: 12-20.

Lacasta D et al., 2021. Epidemiological study related to the first outbreak of ovine anaplasmosis in Spain. Animals 11: Article # 2036.

Li H et al., 2015. Human infection with a novel tick-borne Anaplasma species in China: a surveillance study. The Lancet Infectious Diseases 15: 663-670.

Madison-Antenucci S, 2020. Emerging tick-borne diseases. Clinical Microbiology Reviews 33: e00083-18.

Maggi RG and Krämer F, 2019. A review on the occurrence of companion vector-borne diseases in pet animals in Latin America. Parasites and Vectors 12: 1-37.

Maggi RG et al., 2013. Co-infection with *Anaplasma platys*, *Bartonella henselae* and Candidatus *Mycoplasma haematoparvum* in a veterinarian. Parasites and Vectors 6: 1-10.

Manickam R, 1987. Epidemiological and clinical observations of acute anaplasmosis in sheep. Indian Journal of Veterinary Medicine 7: 159-60.

Martin AR et al., 2005. Anaplasma platys: an improved PCR for its detection in dogs. Experimental Parasitology 109: 176-180.

Melo AL et al., 2016. A survey of tick-borne pathogens in dogs and their ticks in the Pantanal Biome, Brazil. Medical and Veterinary Entomology 30: 112-116.

Palmer GH, 2001. Strain composition of the Ehrlichia, Anaplasma marginale within persistently infected cattle, a mammalian reservoir for tick transmission. Journal of Clinical Microbiology 39: 631-635.

Palomar AM et al., 2015. Detection of tick-borne *Anaplasma bovis, Anaplasma phagocytophilum* and *Anaplasma centrale* in Spain. Medical and Veterinary Entomology 29: 349-353.

Parola P et al., 2003. Detection of Ehrlichia spp., Anaplasma spp., Rickettsia spp., and other eubacteria in ticks from the Thai-Myanmar border and Vietnam. Journal of Clinical Microbiology 41: 1600-1608.

Paterson T et al., 2020. Heartworm control in Grenada, West Indies: Results of a field study using imidacloprid 10%+ moxidectin 2.5% and doxycycline for naturally-acquired Dirofilaria immitis infections. Veterinary Parasitology 284: Article # 109194.

Paulauskas A et al., 2012. Molecular detection and characterization of *Anaplasma phagocytophilum* strains. Comparative Immunology, Microbiology and Infectious Diseases 35: 187-195.

Peng Y et al., 2021. The first detection of *Anaplasma capra*, an emerging zoonotic Anaplasma sp., in erythrocytes. Emerging Microbes and Infections 10: 226-234.

Piratae S et al., 2019. Molecular evidence of *Ehrlichia canis* and *Anaplasma platys* and the association of infections with hematological responses in naturally infected dogs in Kalasin, Thailand. Veterinary World 12: 131-135.

Primo ME et al., 2019. Development of a novel fusion protein with *Anaplasma marginale* and *A. centrale* MSP5 improved performance of Anaplasma antibody detection by cELISA in infected and vaccinated cattle. PLoS ONE 14: e0211149.

Qurollo BA et al., 2014. Co-infection with *Anaplasma platys*, *Bartonella henselae*, Bartonella koehlerae and 'Candidatus *Mycoplasma haemominutum'* in a cat diagnosed with splenic plasma cytosis and multiple myeloma. Journal of Feline Medicine and Surgery 16: 713-720.

Raboloko OO et al., 2020. Seroprevalence of selected tick borne pathogens and diversity and abundance of ixodid ticks (Acari: Ixodidae) at the wildlife-livestock interface in Northern Botswana. Frontiers in Veterinary Science 7: 187-104.

Rajput ZI et al., 2005. Comparative study of *Anaplasma parasites* in tick carrying buffaloes and cattle. Journal of Zhejiang University. Science B 6: 1057-1062.

Rar V and Golovljova I, 2011. Anaplasma, Ehrlichia and "*Candidatus neoehrlichia"* bacteria: pathogenicity, biodiversity and molecular genetic characteristics, a 735 review. Infection, Genetics and Evolution 11: 1842-1861

Richey EJ, 1981. Management of anaplasmosis in beef cattle. The Bovine Practitioner 16: 134-136.

Rocco JM et al., 2020. Severe anaplasmosis represents a treatable cause of secondary hemophagocytic lymphohistiocytosis: Two cases and review of literature. Ticks and Tick-Borne Diseases 11: Article # 101468.

Rochlin I and Toledo A, 2020. Emerging tick-borne pathogens of public health importance: a mini-review. Journal of Medical Microbiology 69: 781-791.

Rodino KG et al., 2020. Tick-borne diseases in the United States. Clinical Chemistry 66: 537-548.

Rodrıguez SD et al., 2000. *Anaplasma marginale* inactivated vaccine: dose titration against a homologous challenge. Comparative Immunology, Microbiology and Infectious Diseases 23: 239-252.

Saetiew N et al., 2020. Spatial and seasonal variation in the prevalence of *Anaplasma marginale* among beef cattle in previously flooded regions of Thailand. Agriculture and Natural Resources 54: 355-362.

Sainz Á et al., 2015. Guideline for veterinary practitioners on canine ehrlichiosis and anaplasmosis in Europe. Parasites and Vectors 8: 1-20.

Sanogo YO et al., 2003. First evidence of *Anaplasma platys* in *Rhipicephalus sanguineus* (Acari: Ixodida) collected from dogs in Africa. Onderstepoort Journal of Veterinary Research 70: 205-12.

Santos AS et al., 2004. Detection of *Anaplasma phagocytophilum* DNA in Ixodes ticks (Acari: Ixodidae) from Madeira Island and Setubal district, mainland Portugal. Emerging Infectious Diseases 10: 1643-1648.

Santos CF and Carvalho CB, 2006. First report of *Anaplasma bovis* at micro region of Campos dos Goytacazes, State of Rio de Janeiro, Brazil. Revista brasileira de parasitologia veterinaria Brazilian journal of veterinary parasitology: Orgao Oficial do Colegio Brasileiro de Parasitologia Veterinaria 15: 126-127.

Sarangi LN et al., 2021. Prevalence of antibodies to Anaplasma in cattle and buffaloes of different organized herds in India. Journal of Parasitic Diseases 45: 359-365.

Sazmand A et al., 2020. Parasitic diseases of equids in Iran (1931–2020): a literature review. Parasites and Vectors 13: 1-19.

Shompole S et al., 1989. Cloned DNA probes identify *Anaplasma ovis* in goats and reveal a high prevalence of infection. Journal of Clinical Microbiology 27: 2730-2735.

Siddiki AZ et al., 2010. Coproscopic and Haematological Approaches to Determine the Prevalence of Helminthiasis and Protozoan Diseases of Red Chittagong Cattle (RCC) Breed in Bangladesh. Pakistan Veterinary Journal 30: 1-6.

Smith RD et al., 1986. Isolate of Anaplasma marginale not transmitted by ticks. American Journal of Veterinary Research 47: 127-129.

Solomon A and Tanga BM, 2020. The First investigation of tick vectors and tick-borne diseases in extensively managed Cattle in Alle district, Southwestern Ethiopia. Veterinary Medicine International 19: Article # 8862289.

Sparagano OA et al., 2003. Molecular detection of Anaplasma platys in dogs using polymerase chain reaction and reverse line blot hybridization. Journal of Veterinary Diagnostic Investigation 15: 527-534.

Splitter EJ et al., 1955. Anaplasmosis in sheep in the United State. Journal of the American Veterinary Medical Association 127: 244–245.

Stich R et al., 1989. Transstadial and attempted transovarial transmission of *Anaplasma marginale* by Dermacentor variabilis. American Journal of Veterinary Research 50: 1377-1380.

Stuen S et al., 2002. Reduced weight gain due to subclinical *Anaplasma phagocytophilum* (formerly Ehrlichia phagocytophila) infection. Experimental and Applied Acarology 28: 209-215.

Stuen S et al., 2018. Intrauterine transmission of *Anaplasma phagocytophilum* in persistently infected lambs. Veterinary Sciences 5: 25-33.

Suksawat J et al., 2001. Serologic and molecular evidence of coinfection with multiple vector-borne pathogens in dogs from Thailand. Journal of Veterinary Internal Medicine 15: 453-462.

Teshale S et al., 2015. Improved molecular detection of Ehrlichia and Anaplasma species applied to Amblyomma ticks collected from cattle and sheep in Ethiopia. Ticks and Tick-Borne Diseases 6: 1-7.

Tomanović S et al., 2013. Analysis of pathogen co-occurrence in host-seeking adult hard ticks from Serbia. Experimental and Applied Acarology 59: 367-376.

Tumwebaze MA et al., 2020. First molecular detection of Babesia ovis, Theileria spp., Anaplasma spp., and Ehrlichia ruminantium in goats from western Uganda. Pathogens 9: 895-104.

Uilenberg G, 1995. International collaborative research: significance of tick-borne hemoparasitic diseases to world animal health. Veterinary Parasitology 57: 19-41.

Uilenberg G, 1997. General review of tick-borne diseases of sheep and goats world-wide. Parasitologia 39: 161-165.

Villar M et al., 2016. The intracellular bacterium *Anaplasma phagocytophilum* selectively manipulates the levels of vertebrate host proteins in the tick vector Ixodes scapularis. Parasites and Vectors 9: 1-17.

Waruri SK et al., 2021. Bovine Anaplasmosis and Control. In Combating and Controlling Nagana and Tick-Borne Diseases in Livestock. IGI Global pp: 221-242.

Wickwire KB et al., 1987. Infectivity of three *Anaplasma marginale* isolates for Dermacentor andersoni. American Journal of Veterinary Research 48: 96-99.

Woldehiwet Z, 2006. *Anaplasma phagocytophilum* in ruminants in Europe. Annals of the New York Academy of Sciences 1078: 446-460.

Woldehiwet Z, 2010. The natural history of *Anaplasma phagocytophilum*. Veterinary Parasitology 167: 108-122.

Yasini SP et al., 2012. Hematologic and clinical aspects of experimental ovine anaplasmosis caused by *Anaplasma ovis* in Iran. Iranian Journal of Parasitology 7: 91-98.

Yoshihara E et al., 2003. Studies of natural infection with Anaplasma marginale in nelore cattle in the Umuarama municipality, Parana State, Brazil. Revista Brasileira de Parasitologia Veterinária 12: 21-26.

Yousif YA et al., 1983. Anaemia due to anaplasmosis in Iraqi goats: Clinical and haematological features under field conditions. Indian Veterinary Journal 60: 576-578.

Zaugg JL, 1985. Bovine anaplasmosis: transplacental transmission as it relates to stage of gestation. American Journal of Veterinary Research 46: 570-572.

Zobba R et al., 2020. Molecular epidemiology of Anaplasma spp. related to *A. phagocytophilum* in Mediterranean small ruminants. Acta Tropica 202: Article # 105286.