Chapter

Identification of Ticks in Dogs with Ehrlichiosis

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Abstract

Examination of ticks collected from ehrlichiosis positive dogs revealed the occurrence of *Rhipicephalus sanguineus*. The distribution of ehrlichiosis in dogs is related to the spreading of vectors. *Ehrlichia canis* is the etiologic agent of canine monocytic ehrlichiosis (CME) and recognized as the most prevalent tick-borne disease affecting dogs and is transmitted by the brown dog tick *Rhipicephalus sanguineus* with an expanding global distribution. Infection of the vertebrate host occurred when an infected tick ingested a blood meal which in turn contaminated the feeding site with its salivary secretion. Blood transfusions from infected donors can also transmit the organisms. Hence, identification of ticks is necessary to detect the disease affecting dogs.

Keywords: ticks, ehrlichiosis, Monocytosis, canine, *Rhipicephalus sanguineus*

1. Introduction

Ehrlichial diseases have emerged as significant problems for human and animals over the past two decades [1–3]. In 1935, *Ehrlichia canis* was first discovered in dog in Algeria [4]. Before the outbreak in military working dogs in Southeast Asia in 1967, canine ehrlichiosis was considered to be a mild disease characterized by fever, vomiting and naso-ocular discharge [5]. Since then, the disease in dogs has spread worldwide [6] and caused serious effects.

Canine ehrlichiosis or tropical pancytopenia is an acute, subacute or chronic tick borne disease caused by *E. canis* which is a Gram-negative intracellular bacterium [7] which occurs particularly in tropical and subtropical regions due to its geographical distribution of its vector tick *Rhipicephalus sanguineus* [8]. This disease is characterized clinically by anorexia, fever, vomiting, loss of weight, enlargement of the liver, spleen and lymph nodes, epistaxis, superficial bleeding and thrombocytopenia [9]. Dogs with canine monocytic ehrlichiosis may die due to hemorrhage and/or secondary infection [10]. Hematological changes in dogs affected with *E. canis* of all stages of infection include a reduction in hematocrit, hemoglobin concentration, blood cell count, thrombocytopenia and leucopenia. However, poor reticulocyte response (non-regenerative anemia) is associated with chronic ehrlichiosis [11]. Diagnosis of ehrlichiosis can be made based on clinical signs, demonstration of morulae in the monocytes, serological testing with the detection of antibodies against *E. canis* and polymerase chain reaction (PCR). Demonstration of morulae inclusions in blood smears of dogs in the subclinical and chronic stages of the disease was often difficult or impossible and has a low sensitivity rate as this

organism is usually present in very low concentrations and hence cannot be used for diagnosis of the disease [10]. The evolutionary thesis suggests that both ixodid and argasid ticks have been in existence since the late Paleozoic to early Mesozoic eras [12]. Antecedent forms evolved as obligate ectoparasites of smooth-skinned reptiles during the late Paleozoic era [5].

Ticks are the most important ectoparasites in tropical and sub-tropical areas. They are also responsible for severe economic losses either through direct effects of blood sucking or indirectly as vectors of pathogens and toxins. Ticks (Acari: Ixodida) are blood feeding ectoparasites acts as vectors of human diseases next to mosquitoes, but comparatively more important as vectors of animal diseases [13–15]. Ticks belong to

Phylum: Arthropoda
Class: Arachnida
Subclass: Acari
Order: Parasitiformes
Suborder: Ixodida

Ixodida contains three families: Argasidae (soft ticks having dorsum without chitin), Ixodidae (hard ticks having dorsum totally or partially covered with chitin) and Nuttalliellidae (an ill known monotypic family represented by Nuttalliella namaqua), among which Argasidae and Ixodidae are more important. In turn, according to morphological characters, the family Ixodidae is subdivided into the Prostriata group (genus Ixodes) and Metastriata group (all other genera in Ixodidae).

Traditionally, classifications and phylogenetics inferences for Ixodida were based on morphological, biological and ecological characteristics [16–21]. Tick classification largely based on morphological characteristics, and the value given to differences and similarities among groups of ticks, resulting in non-homogeneous tick arrangements. The molecular taxonomy associated with conventional morphological cataloging will be useful to obtain a more homogeneous and independent criterion for classification, although in the short term this may not be obvious.

1.1 Importance of tick identification in dogs

Many ticks are responsible for causing various diseases. Among which the tick *Rhipicephalus sanguineus* (brown dog tick) plays vital role because it causes ehrlichiosis in dogs which is life threatening disease in dogs which causes symptoms similar to that of dengue in human beings. Reduction in platelet count and multi-organ failure are the major detrimental things in case of dogs in this specific ehrlichiosis disease. That is why identification of ticks is very important to rule out the disease and early identification will help in saving the life of the animal by giving appropriate treatment. Presence of ticks itself will help in identifying subacute cases so that life loss can be avoided. Due to its veterinary and public health relevance, *Rhipicephalus sanguineus* is one of the most studied ticks.

Ticks able to survive in adverse conditions too as they have heavy protective, chitinous covering and can withstand long periods of starvation and also have wide host range. They can deposit large number of eggs at a time and are relatively free from natural enemies and are tenacious blood suckers.

Medical and veterinary importance of ticks based on their capability of disease transmission. The important diseases transmitted by ticks are Lyme borreliosis (*Borrelia burgdorferi*), *Canine babesiosis* (*babesia* sp.), Ehrlichiosis (*Ehrlichia* sp.), Anaplasmosis (*Anaplasma* sp.), Hepatozoonosis (*Hepatozoon* sp.).

2. Materials and methods

The dogs presented to Teaching Veterinary Clinical Campus that were diagnosed for ehrlichiosis by nPCR were utilized for the study. Around 3 or 4 ticks collected from different sites of the affected dogs were fixed in a 70% ethanol solution. It was further processed and was identified as per the morphology described by [14]

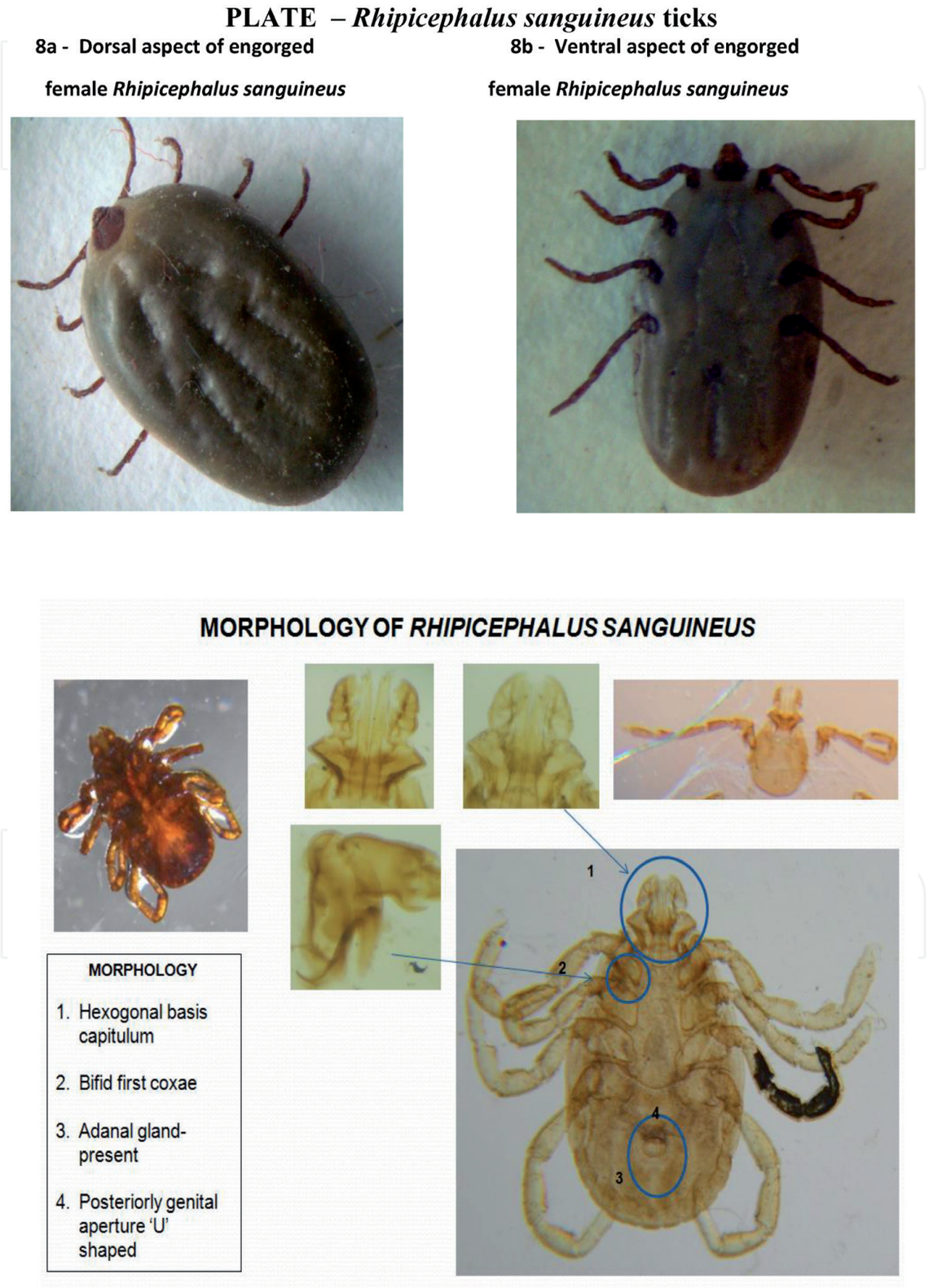

Figure 1.
Rhipicephalus sanguineus *ticks.*

using stereomicroscope and magnifying lens. The stereomicroscope was used at a low magnification of 10× and magnifying lens was used at 40× magnification for identification of specific morphological features.

2.1 Collection of ticks

Unengorged/engorged male and female ticks were collected from dogs either by gently plucking from the body of the dog by hand manipulation or with the help of blunt pointed forceps without damaging their mouth parts. The specimen collected in a plastic container with ventilated cap was labeled appropriately as per host and sites of attachment. Label must contain information about date and place of collection, host, age and site of collection. These samples were transported to the laboratory for further studies.

2.2 Tick identification

These ticks were identified using standard keys [22, 23].

The ticks in the present study were identified as *R. sanguineus* (**Figure 1**). Sen and Fletcher [24] reported that *R. sanguineus* was the only tick that infested dogs in India. Bashir et al. [25] from Pakistan reported 96.8% of the ticks were identified as *R. sanguineus* and the remaining identified as *Dermacentor* and *Haemaphysalis* species. In the present study, all the ticks were identified as *R. sanguineus* and concurred with the findings of [24]. Krogt [26] demonstrated that *R. sanguineus* ticks were able to transmit *E. canis* from a naturally infected dog to an uninfected dog via the bite of the infected tick. Filippova [27] from Japan reported that *E. canis* developed in the salivary glands of *R. sanguineus.* Though, *R. sanguineus* seems to be the vector for *E. canis* in Puducherry, definite studies regarding tick transmission of ehrlichiosis caused by *E. canis* in India is lacking. Hence, transmission studies needs to be undertaken to determine its vector potentiality.

2.3 Tick control measures

Economic losses can be reduced by adopting tick control measures like chemical acaricides [7]. The major reason to control ticks includes disease transmission, tick paralysis or toxicosis by *Rhipicephalus* sp. [7] and physical damage caused by ticks. Keeping animals away from tick-prone areas is the most effective way to control exposure.

3. Results and discussion

Out of 46 dogs found positive for ehrlichiosis, 35 dogs (76.10%) were infested with ticks (**Figure 2**). The ticks collected from different sites of the dogs suffering from ehrlichiosis were identified as *R. sanguineus* based on specific morphological features viz. the reddish brown scutum and conscutum, slightly convex shaped eyes, hexagonal basis capitulum, bifid first coxae, posterior "U" shaped genital aperture and the presence of adanal glands [9]. Bashir et al. [25] from Pakistan reported 96.8% of the ticks were identified as *R. sanguineus* and the remaining identified as *Dermacentor* and *Haemaphysalis* species. *R. sanguineus* was the most commonly encountered tick in India as reported by [28]. In the present study, all the ticks were identified as *R. sanguineus* which concurred with the findings of [24] who reported that *R. sanguineus* was the only tick that infested dogs in India [29]. Filippova [27] reported that *E. canis* developed in the salivary glands of *R. sanguineus* and were able

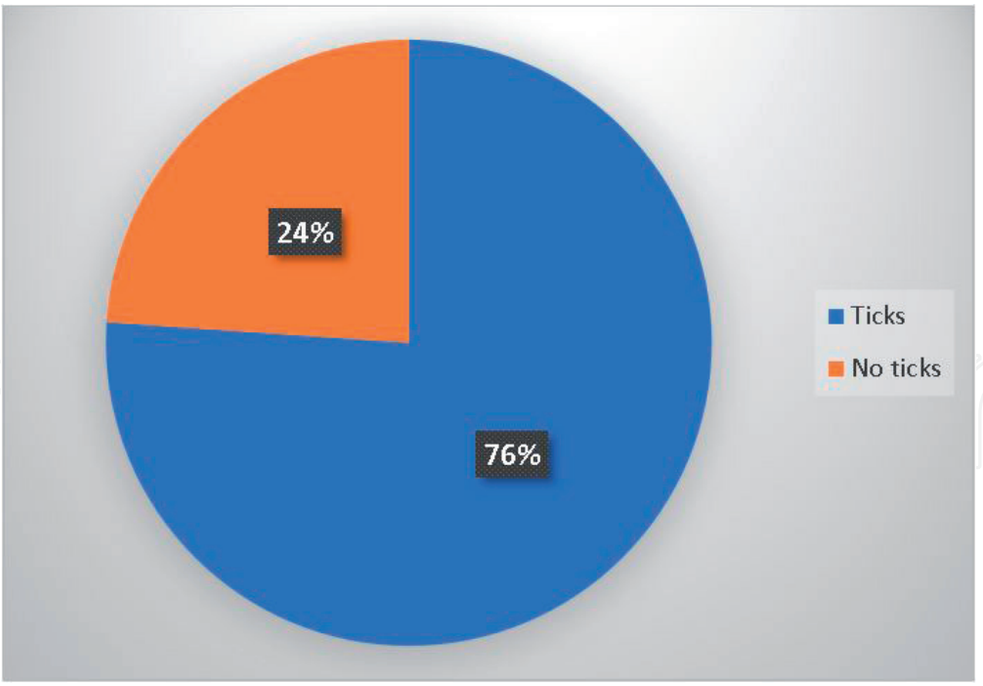

Figure 2.
Representation of ticks in dogs with ehrlichiosis.

to transmit *E. canis* from a naturally infected dog to an uninfected dog via the bite of an infected tick. Although, *R. sanguineus* seems to be the vector for spread of *E. canis*, definite studies regarding tick transmission of the disease is lacking in India. Hence, transmission studies needs to be undertaken to determine its vector potentiality.

4. Summary

The present study on ticks collected from 35 dogs affected with *Ehrlichia canis* were identified as *R. sanguineus* based on the typical morphological features which included hexagonal basis capitulum, bifid first coxae, presence of adanal shields, posterior "U" shaped genital aperture and the presence of adanal glands. Hence, it is concluded that *R. sanguineus* ticks were responsible for transmitting *E. canis* infection in dogs of Puducherry.

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