

Serological Survey of *Rickettsia*, *Anaplasma*, *Ehrlichia*, *Babesia*, and Flaviviruses in Dogs from Western Sicily (Italy)

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INTRODUCTION & AIM

Tick-borne diseases (TBDs) are increasingly recognized as significant emerging and re-emerging zoonotic threats worldwide. These infections are caused by a wide range of pathogens, including bacteria such as *Rickettsia* spp., *Ehrlichia* spp. [1], and *Anaplasma* spp.; protozoa such as *Babesia* spp.; and viruses belonging to the *Flaviviridae* family, including the Tick-Borne Encephalitis Virus (TBEV).

In Mediterranean regions, favorable environmental and climatic conditions support the survival and reproduction of tick vectors, thereby facilitating the maintenance and spread of pathogens. Sicily (Italy), characterized by a mild climate and ecological diversity, represents a potentially endemic area for several of these vector-borne agents.

Dogs, because of their close contact with humans and exposure to similar environmental risks, serve as effective sentinels for monitoring the circulation of TBDs and for evaluating their zoonotic potential.

The aim of this study was to determine the seroprevalence of *Rickettsia* spp., *Ehrlichia* spp., *Anaplasma* spp., *Babesia* spp., and flaviviruses in dogs from western Sicily. The study provides updated epidemiological data that are relevant to both veterinary and public health, contributing to a better understanding of TBDs circulation within a One Health framework (Figure 1).

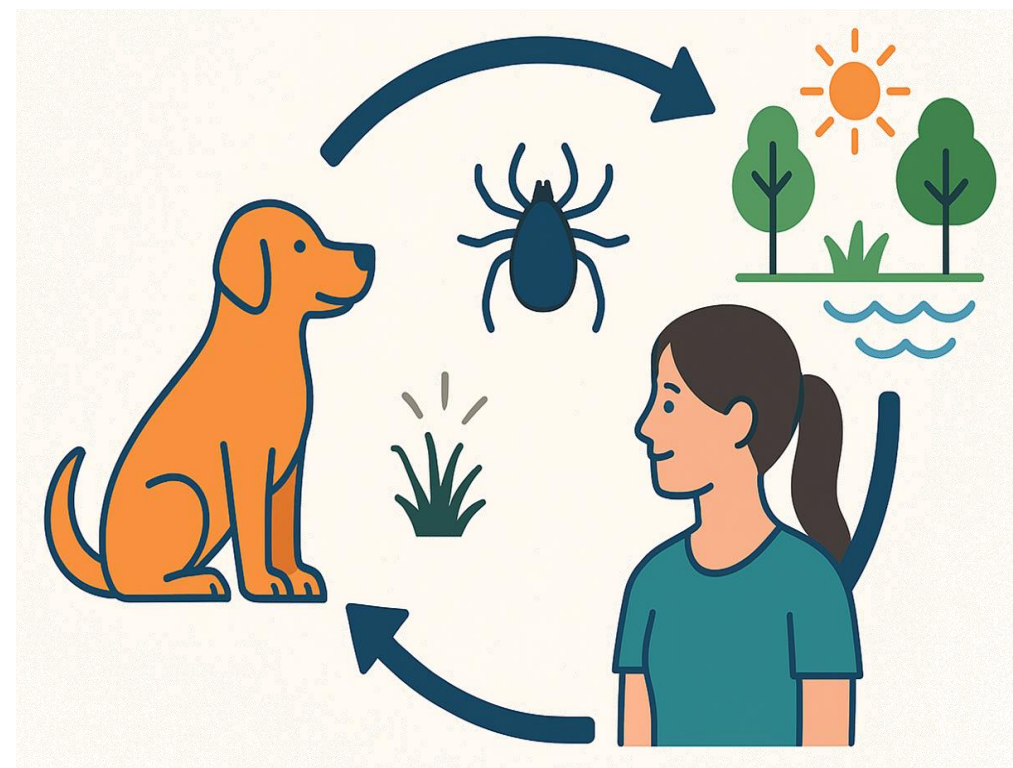


Figure 1. Tick-borne pathogens: a One Health perspective

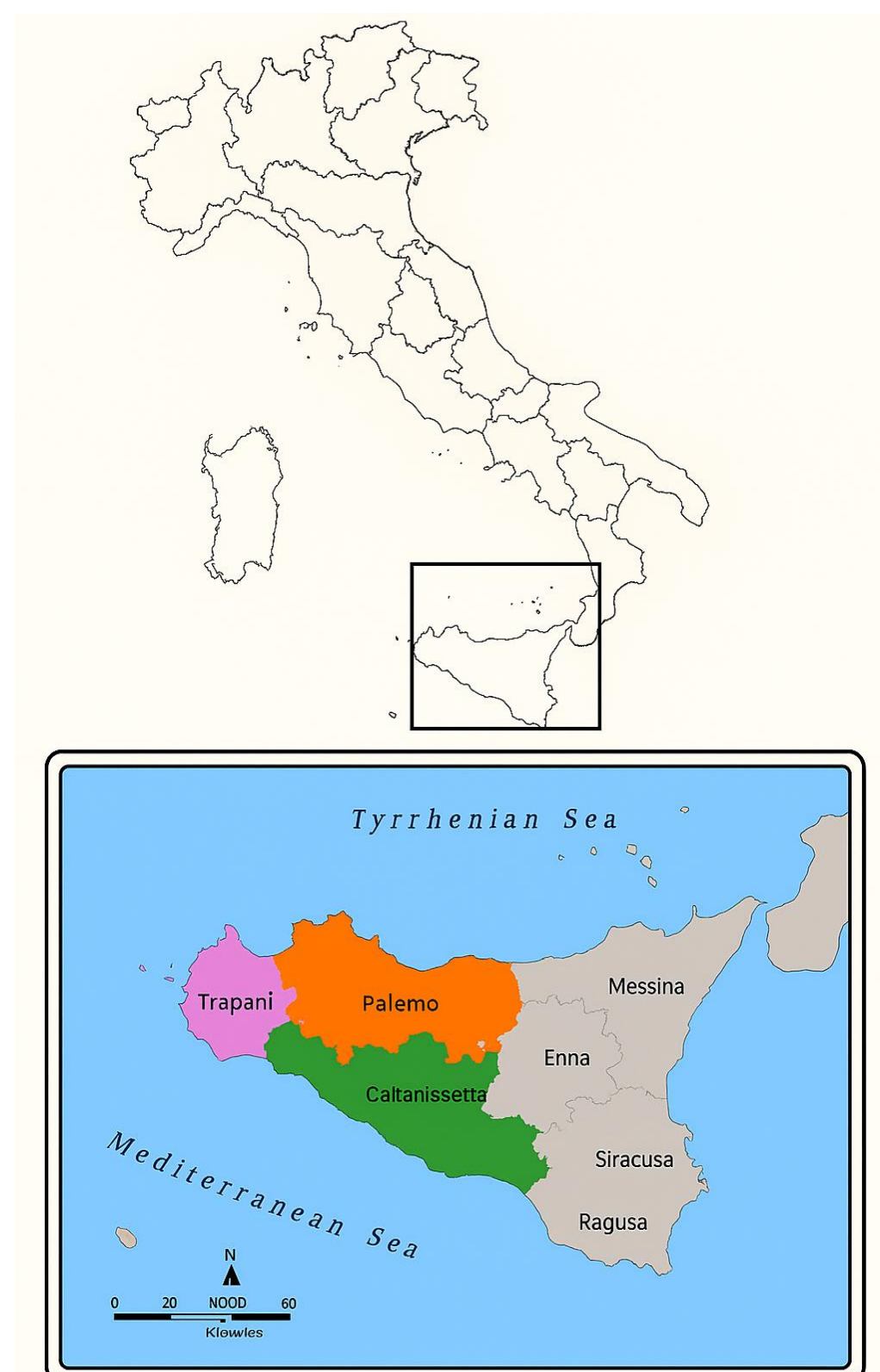


Figure 2. Map of Sicily (Italy) showing the sampling areas. The colored provinces (Palermo, Trapani, and Agrigento) indicate the regions where samples were collected for serological analysis.

METHODS

Blood samples were collected from stray dogs originating from various provinces of western Sicily, specifically Palermo, Trapani, and Agrigento (Figure 2), encompassing both urban and rural areas. Since the animals were free-ranging and unowned, their clinical history and health status at the time of sampling were unknown.

Serological analyses were performed to detect antibodies against tick-borne pathogens. Indirect Immunofluorescence Assay (IFA) was used to identify antibodies against *Rickettsia* spp., *Anaplasma* spp., and *Ehrlichia* spp., employing commercial kits (Fuller Laboratories) and following the manufacturer's instructions. Enzyme-Linked Immunosorbent Assay (ELISA) was used to detect antibodies against *Babesia* spp. and TBEV with Gold Standard Diagnostics kits. It should be noted that the TBEV ELISA test is not specific for TBEV, as it may cross-react with other flaviviruses; therefore, positive results should be interpreted as evidence of exposure to flavivirus antigens rather than a definitive indication of TBEV infection. All samples were processed and interpreted according to the manufacturer's protocols. Descriptive statistical analyses were conducted to determine overall and pathogen-specific seroprevalence rates, providing an overview of the exposure patterns to tick-borne pathogens in the canine population.



RESULTS & DISCUSSION

A total of 386 canine blood samples were analyzed to assess exposure to major tick-borne pathogens in western Sicily (Figure 2). Serological testing revealed a high circulation of bacterial agents. In particular, antibodies against *Rickettsia* spp. were detected in 47.1% of the samples, indicating that nearly half of the dogs had been exposed to rickettsial agents. Antibodies against *Anaplasma* spp. and *Ehrlichia* spp. were found in 21.3% and 15.5% of samples, respectively, confirming the widespread presence of these pathogens in the study area.

Regarding protozoan infections, ELISA testing showed a seropositivity for *Babesia* spp. of 32.4%, highlighting a significant circulation of this parasite among the canine population. Additionally, a subset of 121 samples (approximately 30% of the total) was screened for antibodies against flaviviruses, revealing a seroprevalence of 26.4%. This finding suggests exposure to viruses within this group, possibly including TBEV, although confirmation requires additional serum neutralization testing (Figure 3).

A high frequency of co-exposure to multiple pathogens was observed, reflecting the complex dynamics of vector-borne diseases and the presence of environmental conditions favorable to tick survival and pathogen transmission. Overall, the results confirm the active circulation of *Rickettsia* spp., *Anaplasma* spp., *Ehrlichia* spp., *Babesia* spp., and flaviviruses in western Sicily.

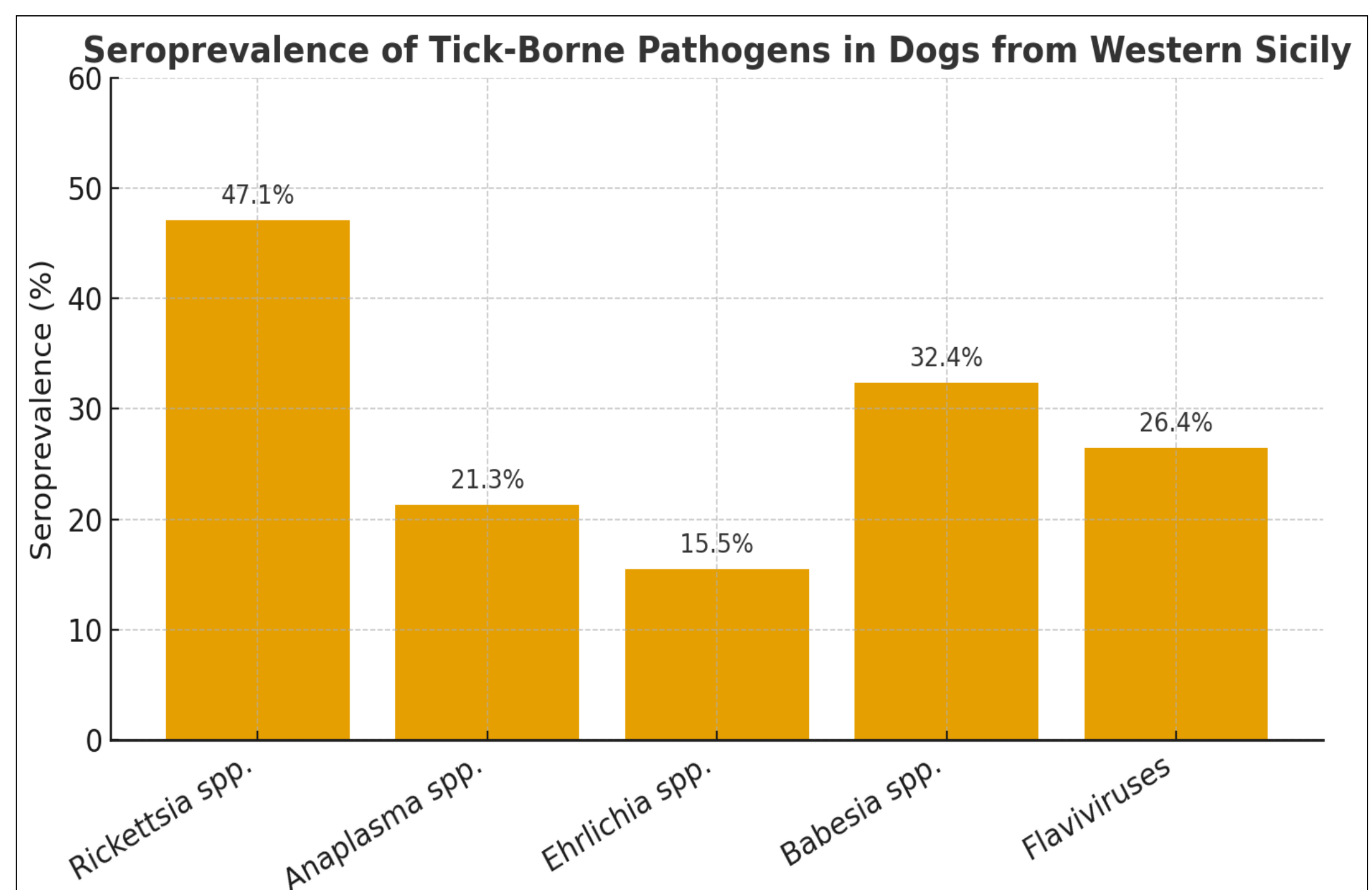


Figure 3. Seroprevalence of tick-borne pathogens in dogs from western Sicily. Bar chart showing the percentage of canine sera positive for antibodies against *Rickettsia* spp., *Anaplasma* spp., *Ehrlichia* spp., *Babesia* spp., and flaviviruses.

CONCLUSION

Within a One Health perspective, dogs represent valuable sentinel hosts for zoonotic infections [2,3], as their exposure patterns closely mirror those of human populations sharing the same environment. These findings highlight the importance of continuous epidemiological surveillance aimed at early detection of emerging pathogens and at guiding effective control strategies.

The high seroprevalence observed confirms the intense circulation of multiple tick-borne pathogens in the region. Continuous surveillance programs, combined with tick control measures, public health awareness, and vaccination strategies where available, are essential to reduce the risk of transmission. An integrated One Health approach is crucial to protect both animal and human health in Mediterranean ecosystems.

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