This study reports a serological and molecular survey on the spotted fever group Rickettsia found in a group of 51 dogs with suspected tickborne illness from the south of Portugal. Additionally, a prevalence of IgG antibodies to R. conorii was also estimated in a group of 400 healthy dogs from the same region. In the group of healthy dogs, the immunofluorescence test revealed that 154 (38.5%) of the 400 dogs had IgG antibodies reactive with R. conorii. The highest proportion of dogs (45%) with R. conorii antibodies was found in blood samples collected from October to December. Among the group suspected with tickborne illness, 35 (62%) dogs showed to be seroreactive (IgG≥128) for antibodies against R. conorii, and the analysis of PCRpositive amplicons revealed that 5 dogs were infected with R. conorii Malish and 2 dogs were infected with R. conorii Israeli tick typhus strain. There was a higher prevalence of antibodies to R. conorii in the group of sick dogs, and the detection of R. conorii DNA in blood samples from this group points to their potential role as a reservoir and sentinel host helping to evaluate and characterize the distribution of circulating rickettsial strains.