

Komunikaty

Results of study over *Ehrlichia canis* appearing in ticks infesting dogs in Lower Silesian Region*

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ABSTRACT. *Ehrichia canis* is an etiologic agent of canine monocytic ehrlichiosis — usually asymptomatic in early stage of disease. It is a tick-borne disease. The aim our study was to estimate the prevalence of ricketsia in the tick population, that infested dogs in Lower Silesia region during season 2005 by PCR. The study took to consideration of identification of ticks species and possibility of season-influence on appear of infections. Among collected ticks (n=102), 97% represented *Ixodes ricinus*. Other species infesting dogs were *Ixodes hexagonus* (n=1) and *Hemaphysalis punctata* (n=2). All examined ticks were negative in the PCR reaction of *Ehrlichia canis*. Because of negative results there was no possibility to establish season-influence of appearing ricketsia infections.

Key words: *Ehrlichia canis*, PCR, ticks.

Introduction

Canine Monocytic Ehrlichiosis (CME) is a ticks-transmitted dog disease caused by *Ehrlichia canis*. Early symptoms of CME are very often similar to flu symptoms and are most likely to be unnoticed by the owner. We have shown that positive titers ranging from 1:20 to 1:1280 for *Ehrlichia canis* specific antibody exist in dogs from Lower Silesian Region [1].

This study has been established to further estimate prevalence of ricketsia in ticks infesting dogs from the same region.

Material and methods

102 specimes representing three ticks species: *Ixodes ricinus* (n=99), *Haemaphysalis punctata* (n=2) *i Ixodes hexagonus* (n=1) have been collected from 102 dogs treated in veterinary clinics from May to November of 2005.

Nested-PCR technology has been used to investigate presence of *Ehrlichia canis* DNA in collected ticks specimens. QIAamp (r) DNA Tissue Kits [QIAGEN Inc. Syngen Biotech] with Baumgarten modification have been used for DNA extraction [2]. 50 µl of volume has been used to perform the reaction. The following primers used in this experiment have been recommended by dr S. Harrus, the Hebrew University of Jerusalem, Koret Scholl of Veterinary Hospital, Rehovot, Israel:

ECC (5'-AGAACGAACGCTGGCGGCAAGCC-3'),
ECB (5'-CGTATTACCGCGGCTGCTGGCA-3'),
HE3 (5'-TATAGGTACCGTCATTATCTTCCCTAT-3'),
"CANIS" (5'-CAATTATTTATAGCCTCTGGC-TATAGGA-3')

DNA extracted from canine monocythes cell line DH 82 infected with *Ehrlichia canis* has been used as positive control. Buffer solution used to perform experimental reaction has been used as a negative control. The expected molecular mass of amplification product was 390 bp. 1.5% agarose gel with ethidium bromide was used to detect the amplificated product of gene 16S rRNA.

Results

We have not detected any amplification products of gene 16S rRNA therefore we conclude that the analysed specimens were not infected with *Ehrlichia canis* (Fig. 1).

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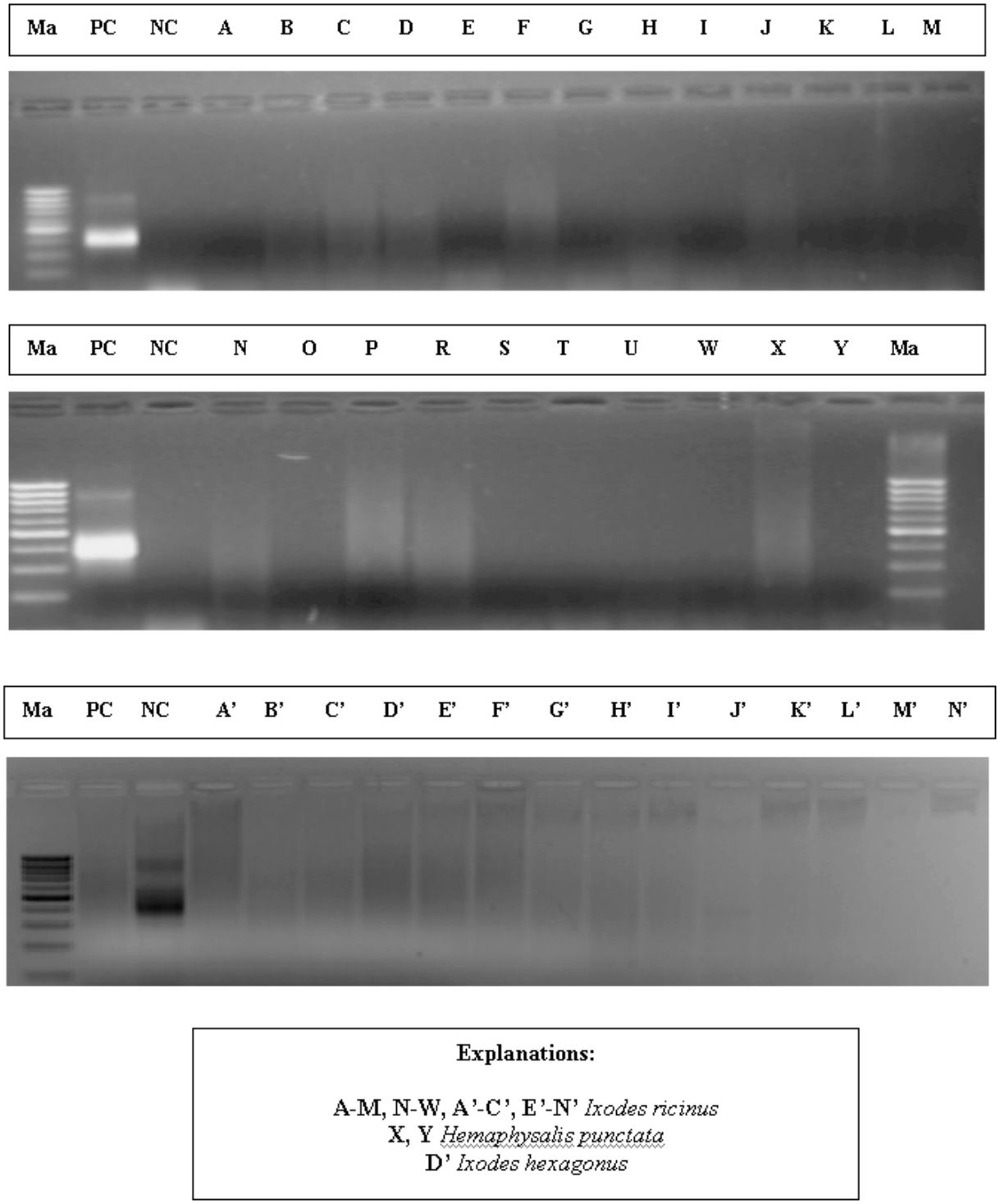

Fig. 1. Agarose gel of *Ehrlichia canis* gen 16S rRNA amplification products. A-X, A'-N'/ examined probes — isolates of ticks DNA, considerated ticks species that infested dogs during season 2005/. Ma — molecular mass marker/"step lader" (100 bp) [SIGMA]. PC — positiv control, NC — negativ control

References

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[2] Baumgarten B.U., Rollinghoff M., Bogdan C. 1999. Prevalence of *Borelia burgdorferi* and granulocytic and monocytic ehrlichiae in *Ixodes ricinus* ticks from southern Germany *Journal of Clinical Microbiology* 37: 3348-3451.

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