Canine ehrlichiosis in Egypt: Sero-epidemiological survey

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ABSTRACT

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A total of 374 dogs, 252 from five military kennels and 122 privately owned, were tested for *Ehrlichia canis* antibody. Sera were tested at a 1:20 dilution by indirect fluorescent antibody with the use of *E. canis* cell-culture antigen slides. The overall prevalence of *E. canis* antibody was 33%. Antibody prevalence among military dogs (29%) was significantly lower than among privately owned dogs (41%; P < 0.05). The *E. canis* seroprevalence among dogs infested with ticks (*Rhipicephalus sanguineus*) was higher (44%) than that among uninfested dogs (31%; P = 0.08). The seroprevalence among military dogs varied from 21–46% at the five kennels; lower prevalences were observed in kennels with higher sanitary and hygienic conditions. Age- and sex-related *E. canis* antibody prevalences were ot significantly different among military and privately owned dogs, although adult and male privately owned dogs had the highest seroprevalences (45% and 44%, respectively). Three dogs with epistaxis had *E. canis* antibody titres > 1:320. These data demonstrate the first laboratory evidence of *E. canis* infection among dogs in Egypt.

Keywords: Ehrlichia, dogs, Egypt, sero-epidemiological survey

INTRODUCTION

Tropical canine pancytopenia (TCP) or canine ehrlichiosis is a tick-borne haemorrhagic disease of dogs caused by the rickettsial organism, *Ehrlichia canis*. The disease is transmitted by the brown dog tick *Rhipicephalus sanguineus* (Groves, Dennis, Amyx & Huxsoll 1975), and is known to occur in Africa, south-east Asia, and the USA. In Africa, the disease was first recognized in Algeria (Donatien & Lestoquard 1935) and later reported in Tunisia (Bobin, Chabassol, De Brux, Fiehrer, Guillot, Michel & Pigoury 1962). The rate of

infection is especially high in tropical zones (up to 30%), and was reported in Puerto Rico (Huxsoll, Hildbrandt, Nims, Ferguson & Walker 1969), the Middle East (Seamer & Snape 1970), and East Africa (Kaminjalo, Nyindo, Sayer, Rurangir, Johnson, Hird, Rosenbaum, Maxie & Ogaa 1976). Recently, 34 of 68 (50%) military dogs from Tunisia, Senegal and Chad were found positive for *E. canis* antibody (Brouqui, Davoust, Haddad, Vidor, Raoult 1991).

Clinically, the most overt sign of the disease is unilateral or bilateral epistaxis (Ewing 1969; Walker, Rundquist, Taylor, Wilson, Andrews, Barek, Hogge, Huxsoll, Hildbrandt & Nims 1970). Death often occurs 5–7 d after epistaxis. Dogs with TCP usually have a history of a febrile episode two or more months before the onset of epistaxis. Following the febrile episode, the dog may appear normal, but pancytopenia persists and animals become severely debilitated. Death may also be attributed to internal hemorrhages. Dogs that

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survive the episode usually remain carriers of the organism for several months and develop antibodies, but are susceptible to reinfection (Weisiger, Ristic & Huxsoll 1975). In the German shepherd, immunosuppression has been observed, with *E. canis* infection causing serious impact on their efficiency (Nyindo, Huxsoll, Ristic, Kakoma, Brown, Carson & Stephenson 1980).

In Egypt, this disease has not been previously reported. However, epistaxis which is a clinical sign suggestive of this disease, has been recognized among German shepherd dogs in military units (Botros, unpublished data 1992). Also, the tick species (*Rhipicephalus sanguineus*) responsible for transmission is quite common in this geographic area (Morel & Vassiliades 1963). Human infection with *E. canis* or a closely related organism was first described in the USA by Maeda, Markowitz, Harvley, Ristic, Cox & Mcdade (1987). However, genetic-sequence-comparison studies have recently indicated that the agent of human ehrlichiosis, while closely related to *E. canis*, is a new species and has been named *Ehrlichia chaffeensis*. (Anderson, Dawson, Jones & Wilson 1991).

The purpose of this study was to survey dogs, privately owned as well as military working dogs, for evidence of exposure to *E. canis*.

MATERIAL AND METHODS

Samples from military working dogs were collected during the period December 1991 to June 1992, at five locations: War Dogs Unit in Almaza, about 5 km east of Cairo, (n = 35); Presidential Guards Dog Unit in Cairo (n = 109); Transportation and Railway Police Unit in Cairo (n = 74); and Transportation and Railway Police Unit in Alexandria (n = 20). All the dogs at these units were included in the study. Samples from privately owned household dogs (n = 122) were collected at the Peoples' Dispensary for Sick Animals (PDSA), where dogs were brought from various parts of Cairo for treatment or for boarding. Samples were obtained from all the dogs presented to the hospital on a randomly selected day, weekly, during April, May and June 1992.

Age, sex, breed, signs of disease, infestation with ticks and history of epistaxis were recorded at the time of sample collection. Blood was drawn by venipuncture and serum was separated and stored at –20 °C. Serum from 11 dog caretakers who had worked at the PDSA for more than 5 years were submitted for *E. canis* antibody testing.

Sera were tested at 1:20 dilution by standard two-step IFA test (Ristic, Huxsoll, Weisiger, Hildbrandt & Nyindo 1972). *E. canis* cell-culture antigen on 8-well slides (ProtaTek International, Inc. St. Paul, MN) was used. Negative and positive control *E. canis* sera were included with each test.

RESULTS

A total of 374 military and privately owned dogs of both sexes and various ages were enrolled in this study. The results are summarized in Table 1. The majority (88%) of military dogs (n = 252) were of the German shepherd breed while privately owned dogs (n = 122) were of various breeds.

Among the military working dogs, 202 (80 %) were males and 50 (20 %), females. Ninety-six per cent were adults older than one year; the average age was 4,2 years. The overall prevalence of *E. canis* antibody among military dogs was 29 %, with a similar figure in young and adult animals. Prevalence among males was not significantly different from females (Table 1). Prevalence was highest (46%) among war dogs, followed by transportation police dogs in Alexandria (35%), police academy dogs (28%), transportation police dogs in Cairo (23%) and presidential guard dogs (21%).

Of the privately owned dogs, 70 (57%) dogs were males and 52 (43%), females. Sixty-six per cent were adults; the average age was 3,5 years. The overall prevalence of *E. canis* antibody among privately owned dogs was 41%. There were no statistically significant differences between young and adult animals, nor between males and females (Table 1).

Overall, antibody prevalence among privately owned dogs (41%) was significantly higher than among military dogs (29%, P < 0.05) and significantly higher among adults and males (45% v 29% and 44% v 30%, respectively). Of the total number of dogs tested for $E.\ canis$ antibody, 54 dogs (14%) were infested with $R.\ sanguineus$ ticks. Forty-four per cent (24/54) of tick-infested dogs, as compared to 31% (100/320)

TABLE 1 Relationship of age, sex and tick infestation to prevalence of *Ehrlichia canis* antibody in military working and privately owned dog populations in Egypt

Risk group category	Military working dogs No. pos./ No. tested (%)	Privately owned dogs No. pos./ No. tested (%)
Young (age < 1 year) Adult (age > 1 year) Male Female Tick-infested Tick-uninfested All dogs	3/10 (30) 71/242 (29) 60/202 (30) 14/50 (28) 1/1 (100) 73/251 (29) 74/252 (29)	14/42 (33) 36/80 (45) * 31/70 (44) ** 19/52 (37) 23/53 (43) 27/69 (39) 50/122 (41) ***

* P < 0,05 (Chi-square test) for the difference in the number of E. canis-seropositive adult dogs between military dogs (71 of 242) and privately owned dogs (36 of 80)</p>

** P < 0,05 (Chi-square test) for the difference in the number of *E. canis*-seropositive male dogs between military dogs (60 of 202) and privately owned dogs (31 of 70)

*** P < 0,05 (Chi-square test) for the difference in the number of *E. canis*-seropositive animals between military dogs (74 of 252) and privately owned dogs (50 of 122)

of uninfested dogs, were positive for *E. canis* antibody (Table 1), but the difference is not statistically significant (P = 0,08). Three dogs had epistaxis or a history of epistaxis shortly before sample collection, and all were *E. canis*-seropositive with antibody titres that exceeded 1:320. Serum samples collected from the 11 dog caretakers were all antibody-negative to *E. canis*.

DISCUSSION

The lower *E. canis* antibody prevalence among military dogs, as compared to privately owned dogs, could be attributed to better housing conditions and to the regular use of acaricides. Only one of all the military dogs tested, was infested with ticks. Prevalence differences at the various military kennels appeared to correlate with the sanitary conditions at each location. Age- and sex-related prevalence differences within the two dog groups were not significantly different, although privately owned adult and male dogs had the highest prevalences, significantly higher than military adult and male dogs (P < 0,05).

Although only three of the dogs studied had epistaxis or a history of epistaxis, all were *E. canis* positive. Epistaxis is reported as a classic clinical sign of *E. canis* infection (Ewing 1969; Walker *et al.* 1970).

Negative *E. canis* antibody results obtained on the serum samples from the dog caretakers are consistent with previous evidence that *E. canis* is not a human pathogen (Anderson *et al.* 1991) and that dogs are not a reservoir of human ehrlichiosis. In summary, data from this study demonstrated the first laboratory evidence of *E. canis* infection among dog populations in Egypt.

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REFERENCES

- ANDERSON, B.E., DAWSON, J.E., JONES, D.C. & WILSON, K.H. 1991. *Ehrlichia chaffeensis*, a new species associated with human ehrlichiosis. *Journal of Clinical Microbiology*. 29:2838–2842.
- BOBIN, A., CHABASSOL, C., DE BRUX, J., FIEHRER, A., GUIL-LOT, G., MICHEL, C. & PIGOURY, L. 1962. Syndrome hemorrhagique thrombotique thrombocytopenique du chien en Tunisie. *Revue Pathologie Generale*, 62:317–334.
- BROUQUI, P., DAVOUST, B., HADDAD, S., VIDOR, E. & RAOULT, D. 1991. Serological evaluation of *Ehrlichia canis* infections in military dogs in Africa and Reunion Island. *Veterinary Microbiology*, 26:103–105.
- DONATIEN, A. & LESTOQUARD, F. 1935. Existence en Algerie d'une rickettsia du chien. Bulletin de la Societe de Pathologie Exotique, 28:418–419.
- EWING, S.A. 1969. Canine ehrlichiosis, in *Advances in Veterinary Science and Comparative Medicine*. 13, edited by C.A. Brandly & C.E. Cornelius. Academic Press: New York, N.Y.: 331–353.
- GROVES, M.G., DENNIS, G.L., AMYX, H.L. & HUXSOLL, D.L. 1975. Transmission of *Ehrlichia canis* to dogs by ticks. *American Journal Veterinary Research*, 36:937–940.
- HUXSOLL, D.L., HILDEBRANDT, P.K., NIMS, R.M., FERGUSON, J.A. & WALKER, J.S. 1969. *Ehrlichia canis* the causative agent of a hemorrhagic disease of dogs? *Veterinary Record*, 85:587.
- KAMINJALO, J.S. (Jr), NYINDO, M.B.A., SAYER, P.D., RURAN-GIR, W.F., JOHNSON, L.W., HIRD, S.F., ROSENBAUM, E., MAXIE, L.S. & OGAA, J.S. 1976. Identification of *Ehrlichia canis* in East Africa. *Veterinary Record*, 99:434–435.
- MAEDA, K., MARKOWITZ, N., HARVLEY, R.C., RISTIC, M., COX, D. & MCDADE, J.E. 1987. Human infection with *Ehrlichia canis*, a leukocytic rickettsia. *The New England Journal of Medicine*, 316:853–856.
- MOREL, P.C. & VASSILIADES, G. 1963. Les Rhipicephalus du groupe sanguineus: especes africianes (Acariens, Ixodoidea). Revue d'Elevage et de Medecine Veterinaire des Pays Tropicaux, 15:343–386.
- NYINDO, M., HUXSOLL, D.L., RISTIC, M., KAKOMA, I., BROWN, J.L., CARSON, C.A. & STEPHENSON, E.H. 1980. Cell-mediated and humoral immune responses of German shepherd dogs and beagles to experimental injection with *Ehrlichia canis*. *American Journal Veterinary Research*, 41:250–253.
- RISTIC, M., HUXSOLL, D.L., WEISIGER, R.M., HILDBRANDT, P.K. & NYINDO, M.B.A. 1972. Serological diagnosis of tropical canine pancytopenia by indirect immunofluorescence. *Infection and Immunity*, 6:226–231.
- SEAMER, J. & SNAPE, T. 1970. Tropical canine pancytopenia and *Ehrlichia canis* infection. *Veterinary Record*, 86:375.
- WALKER, J.S., RUNDQUIST, J.D., TAYLOR, R., WILSON, B.L., ANDREWS. M.R., BAREK. J., HOGGE, A.L. (Jr) HUXSOLL, D.L., HILDEBRANDT, P.K., & NIMS, R.M. 1970. Clinical and clinicopathologic findings in tropical canine pancytopenia. *Journal American Veterinary Medical Association*, 157:43–55.
- WEISIGER, R.M., RISTIC, M. & HUXSOLL, D.L. 1975. Kinetics of antibody response to *Ehrlichia canis* assayed by the indirect fluorescent antibody method. *American Journal of Veterinary Re*search, 36:689–693.