

Spatial Analysis on the Occurrence of *Pneumocystis carinii* in the Shrew *Notiosorex crawfordi* in Fragmented Landscape in Southern California

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Fungi of the genus *Pneumocystis* are widespread group of pulmonary pathogens infecting immunologically immature or immunocompromised humans and animals. Previous studies have shown significant interspecific differences in the occurrence of *P. carinii* in mammals collected from the wild [4]. The existing data indicates that infection with *P. carinii* is host-species specific but the reservoir of the infection has not yet been determined [7–8]. The transmission of the fungus is known to be airborne, and the fungus has a dormant form that remains infective outside the mammalian host for at least several months [1]. The occurrence of *P. carinii* does not seem to be related to any particular habitat [4,6] but tremendous gaps exist in understanding the epidemiology and ecology of *Pneumocystis* organisms. In the present study, a histochemical technique was used to study the spatial distribution and occurrence of *P. carinii* in the desert shrew, *Notiosorex crawfordi*, in a fragmented landscape of southern California. A previous study [6] showed that the percentage of *P. carinii* infected individuals was higher in *N. crawfordi* than those in other sympatric small mammals.

MATERIALS AND METHODS

The *N. crawfordi* were caught with pitfalls [6] in five study sites (Table 1) in San Diego and Riverside counties (California, USA) from January to May in 1998 and in 1999. The Wild Animal Park (117.01 33.09) study site is within a large inland landscape area of coastal sage scrub and grassland habitats. The University of California Elliott Reserve (117.11 32.89) is a large continuous inland site of chamise chaparral intermixed with elements of coastal sage scrub. Lake Skinner (117.08 33.57) is a large inland site of coastal sage scrub with patches of chamise chaparral. The Point Loma Reserve (117.23 32.67) is a small coastal site of maritime succulent scrub and coastal sage scrub habitats. Camp Pendleton is a large coastal site (117.50 33.34) of chamise chaparral and coastal sage scrub. All study sites are within 100 km of each other. The area of undeveloped landscape around the site was determined by drawing a polygon around the trap arrays of a site using the program Topo USA (DeLorme) that extends to any borders or barriers fragmenting the habitat landscape the arrays are within. The edge index was calculated by estimating the size of intact habitats in each site, and dividing that by the number of 500-m sides in each corresponding site that could be fitted to the site without crossing a road. The trap-success rate was used as a surrogate for population density; it is defined as the number of individuals caught per 100 trap-nights (a trap-night equals one pitfall left open for 24 h). The shrews examined for this study were found dead in pitfall traps. They were frozen until necropsy in spring 2000. On necropsy, sex and age of the animal was recorded, and pieces of lung, heart, liver, spleen and kidney were fixed in 10% buffered formalin to produce standard histological sections which were stained with hematoxylin-eosin and Grocott's modification of Gomori's methenamine silver [2]. The slides were examined by a light microscope at x 400. The intensity of infection was measured as cysts found per lung sections [3]. Chi-square tests were used to analyze the differences in the

occurrence of *P. carinii* between years, sex and age groups. Spearman rank correlations procedure was used to analyze the degree of association between occurrence of *P. carinii*, population density of the host, and three landscape variables (Table 1). Statistix® for Windows software package was used in all analyses.

RESULTS AND DISCUSSION

All animals examined were normal on gross pathologic examination. Cyst forms of *P. carinii* were found only in the lungs of the infected hosts, and the intensities of the infections were low. No histopathological changes were seen around the cysts. There was no difference in the percentage positive between years, sexes or age groups (not shown), and the data were pooled for spatial analysis. It was not possible to study seasonal variation [5] in the occurrence of *P. carinii* because only a few shrews are caught during the hot months during summer and fall in our study areas [6].

We found significant differences in the occurrence of *P. carinii* in *N. crawfordi* between study sites (Table 1). The percentage of *P. carinii* infected *N. crawfordi* was significantly ($r_s=0.95$) higher in sites with high population density of the host compared to sites where host density was low. Similar correlation has previously been found in the vole *Microtus agrestis* [5]. This result suggests that hosts, while not necessarily involved in direct infection of other hosts of the same species, are important in the transmission cycle of *P. carinii* organisms. Reduced immunocompetence associated with high population density stress may also contribute to the positive correlation found between the percentage of infected animals and host density. It is of interest that the percentage of *P. carinii* infected shrews found in this study were similar to those found in Palearctic shrews [4], which maintain much higher population densities, and occupy moister habitats than southern California shrews.

The location of the sampling site (coastal-inland), altitude or habitat type did not show any relation to the occurrence of *P. carinii*, nor did the amount of disturbance, or size of the landscape area surrounding the sampling site. Spatial studies of the genetic diversity, and of other life-cycle forms of *P. carinii* organisms in wild animal populations, are needed to provide further insights into the ecology of these elusive organisms.

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Table 1. The location and geographic variables of the study sites, trap-success rate and the percentage of *P. carinii* infected shrews (%) in each site in 1998-1999. PC+ = the number of infected shrews; N = the number of shrews examined.

Site	Area of the landscape (km ²)	Altitude (m)	Edge index	Trap-success rate	PC +	N	%
Wild Animal Park	397	200	0.75	1.00	11	33	33
U.C. Elliott Reserve	120	190	0.92	0.20	2	23	9
Lake Skinner	103	470	0.1	1.00	8	20	40
Point Loma Reserve	5.5	55	0.38	0.86	10	43	23
Camp Pendleton	>100*	300	0.5*	0.57	0	18	0

* Sample locations were spread throughout the habitat on site which includes a matrix of development impacts. Thus these values are estimates.

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