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Investigations on the biology, epidemiology, pathology, and control of *Tunga penetrans* in Brazil: VII. The importance of animal reservoirs for human infestation

Daniel Pilger • Stefan Schwalfenberg • Jörg Heukelbach • Lars Witt • Heinz Mehlhorn • Norbert Mencke • Adak Khakban • Hermann Feldmeier

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Abstract In Brazil tungiasis is endemic in many resourcepoor communities, where various domestic and sylvatic animals act as reservoirs for this zoonosis. To determine the role of animal reservoirs in human tungiasis, a crosssectional study was performed in a traditional fishing community in northeast Brazil. The human and the animal populations were examined for the presence of embedded sand fleas and the prevalence and the intensity of infestation were correlated. The overall prevalence of tungiasis in humans was 39% (95% CI 34-43%). Of six mammal species present in the village, only cats and dogs were found infested. The prevalence in these animals was 59% (95% CI 50-68%). In households, where infested pet animals were present, a higher percentage of household members had tungiasis (42% [95% CI 30-53%] versus 27% [20–33%], p=0.02), and the intensity of the infestation

D. Pilger · S. Schwalfenberg · L. Witt · A. Khakban ·
H. Feldmeier (⊠)
Institute for Microbiology and Hygiene,
Charité—University of Medicine, Campus Benjamin Franklin,
Hindenburgdamm 27,
12203 Berlin, Germany
e-mail: hermann.feldmeier@charite.de

J. Heukelbach Department of Community Health, School of Medicine, Federal University of Ceará, Fortaleza, Ceará 60430-149, Brazil

H. MehlhornDepartment of Zoology and Parasitology,Heinrich Heine University,40225 Düsseldorf, Germany

N. Mencke

Animal Health Division, Bayer HealthCare AG, 51368 Leverkusen, Germany

was higher (six lesions versus two lesions, p=0.01). The intensity of infestation in animals correlated with the intensity of infestation in humans (rho=0.3, p=0.02). Living in a household with an infested dog or cat led to a 1.6-fold (95% CI 1.1–2.3, p=0.015) increase in the odds for the presence of tungiasis in household members in the bivariate analysis and remained a significant risk factor in the multivariate regression analysis. The study shows that in this impoverished community tungiasis is highly prevalent in humans and domestic animals. In particular, it underlines the importance to include animals in control operation aiming at the reduction of disease occurrence in the human population.

Introduction

Tungiasis is an ectoparasitic skin disease caused by the sand flea Tunga penetrans, in which females penetrate into the epidermis of the host (mainly humans, domestic animals, and rats). There, they hypertrophy, expel hundreds of eggs, and eventually die in situ. The zoonosis is widespread in resource-poor urban and rural communities in sub-Saharan Africa, the Caribbean, and South America, with point prevalences between 20% and 55% in the general population (Ade-Serrano and Ejezie 1981; Chadee 1998; Heukelbach et al. 2001; Muehlen et al. 2003; Wilcke et al. 2002). Although by its nature a self-limiting infestation, tungiasis is actually a debilitating disease associated with considerable morbidity (Heukelbach 2005). Sequels are common and include lymphadenopathy, lymphedema, ulcer, gangrene, and auto-amputation of digits (Feldmeier et al. 2002, 2003). Tetanus is a known risk factor in non-vaccinated individuals (Litvoc et al. 1991; Soria and Capri 1953).

T. penetrans infests a broad range of sylvatic and domestic animals (Cooper 1967; Heukelbach et al. 2004; Rietschel 1989). In urban northeast Brazil, rats (*Rattus rattus*) are an important reservoir (Heukelbach et al. 2004). In a study in a poor neighborhood in Fortaleza, Brazil, 67%, 50%, and 24% of dogs, cats, and rats, respectively, were found infested (Heukelbach et al. 2004). In rural Nigeria, pigs are the animals most frequently affected (Ugbomoiko et al. 2007b).

The older literature provides anecdotal evidence that intruders to an endemic area who had spent a night in a shelter together with domestic animals or rodents became heavily infested (Bonnet 1867; Hoeppli 1963). In a rural community in northeast Brazil, houses built of palm stems without a solid floor, i.e., domiciles similar to the dwellings used by travelers in the past, were associated with high odds for household members to be infested with *T. penetrans* (Muehlen et al. 2006). In a rural community in Nigeria, individuals living in a household with pigs had a higher risk to become infested (Ugbomoiko et al. 2007a).

To assess the importance of domestic animals for human infestation a cross-sectional survey was performed in an endemic community, where humans and animals lived in close contact. The results show that pet animals per se were not a risk factor for the presence of tungiasis in the human population. However, the presence of animals infested with *T. penetrans* in a household increased the infestation risk for humans and the likelihood of a high intensity of infestation.

Materials and methods

Study area and population

The study was performed in the village of Balbino (Cascavél Municipality), Ceará State, northeast Brazil. Balbino is a relatively isolated fishing community surrounded by sand dunes near the Atlantic Ocean. In October 2002, the village was inhabited by 148 families with a total population of 630 inhabitants. The population is poor; the streets are not paved, and most houses are located on rather large compounds surrounded by fences. Only about 75% of the households have electricity. Inhabitants were eligible for the study provided they had spent at least 4 days per week in the village during the last 3 months.

The following animal species existed in the village: cats, dogs, horses, donkeys, cattle, and pigs. Rats (*R. rattus*) had never been observed and other sylvatic rodents were only rarely seen at the compound (Heukelbach, unpublished observation). Most of the animals stroll around without limitations but pigs are kept in small shelters in the backyard of the compound. Dogs are held on the compound

and taken for walks by their owners. Animals were included in the study, when they were born in the village or were kept there for at least 2 months.

Study design

The study took place in October 2002, i.e., in the middle of the dry season, when the prevalence of tungiasis peaks (Heukelbach et al. 2005). During the preparatory phase, contacts were made with community leaders and local health personal, and the objectives of the study were explained. A census of the human and animal population was performed and all houses mapped using a geographic positioning system. During the census, demographic data were collected from household members as well as information on the educational level of the household leader, on household income, and on sanitary infrastructure. In a door-to-door survey, household members and all domestic animals present on the compound were carefully examined for the presence of embedded sand fleas according to a previously established guideline (Eisele et al. 2003). If household members or animals were absent, the household was visited again a couple of days later. In animals, the examination focused on paws, abdomen, and muzzle. The following diagnostic findings were considered to be positive for human as well as animal tungiasis (Eisele et al. 2003): a red-brownish spot with a diameter of 1-3 mm with visible posterior segments of the penetrated flea (early stage), a circular whitish lesion with a diameter of 4-10 mm, and a central black dot presenting the posterior segments (mature stage), black crust surrounded by necrotic tissue (late stage with dead parasite). Typical long-term residuals in the skin, lesions altered through manipulation (such as partially or totally removed fleas, with a characteristic crater-like sore in the skin) and suppurative lesions caused by using nonsterile instruments, were recorded as well.

Ethical considerations

The study protocol was approved by the Ethical Committee of the Cascavel Municipality Board. Only individuals who had given informed oral consent were included in the study. In the case of minors, informed oral consent was obtained by their carers. Individuals with tungiasis were treated surgically. Embedded fleas were extracted with a sterile needle, and the remaining sore was covered with antibiotic ointment to prevent superinfection.

Statistical analysis

Data were entered into Epi Info version 6.04d (CDC, Atlanta, GA, USA), checked for errors which might have

Table 1 Socioeconomic characteristics of the 140 participatinghouseholds in Balbino village (140 households, 597 individuals)

	Total number of households	Percents (%)
Education ^a		
Primary school completed	102	73
Secondary school completed	29	21
Higher education	3	2
Not stated	6	4
Monthly household income ^b		
<1 minimal wage	64	46
1–2 minimal wages	69	49
>2 minimal wages	7	5
Housing		
Plastered adobe	66	47
Crude adobe	51	36
Palm stems	23	16
Type of floor		
Sand	14	9
Stamped clay	29	21
Concrete	97	70
Kitchen		
Stall outside on compound	39	28
Indoors	101	72
Dogs per household		
0	91	64
<2	36	26
2–4	12	9
>4	1	1
Cats per household		
0	92	65
<2	27	19
2–4	19	14
>4	2	2
Households with dogs and cats	26	19

^a Of household leader

^b1 minimum wage≈80 US\$/month

occurred during data entry, and transferred to SPSS 11.04 for Macintosh (SPSS Inc., Chicago, IL, USA) for analysis. The χ^2 and Fisher's exact tests were employed to determine the significance of differences of proportions. The Mann– Whitney *U* test was used to compare the number of lesions in individuals from different groups. Putative risk factors were first assessed by bivariate analysis. Exposure variables with a *p*-value <0.05 were entered into a logistic regression model with stepwise, forward, conditional variable entering. For correlation analysis, the Spearman's rank correlation coefficient was determined.

Results

The target population consisted of 630 individuals with a median age of 27 years (range 1–87 years). There were 304

(48%) males and 326 (52%) females. The median household size was 4.6 persons (range one to eight persons). Table 1 summarizes the socioeconomic characteristics of the study area.

Five hundred ninety-seven individuals met the inclusion criteria, and 490 (77%; 228 males and 262 females) were willing to participate and be examined. The overall point prevalence of tungiasis was 39% (95% CI 34-43%). The median intensity of infestation per infested individual was three lesions (interquartile range 2-7). In total, 188 animals were identified; of these, 153 (81%) were examined (Table 2). In 73 households (52%), at least one dog or cat was present. Out of the six different species of domestic animals living in the village, only dogs and cats were found to be infested with T. penetrans. The overall point prevalence in cats and dogs was 59% (95% CI 50-68%), with a median intensity of infestation of 7.5 lesions (interquartile range 3-14). There was no statistical significant difference in prevalence and intensity of infestation between cats and dogs.

Bivariate analysis demonstrated the following exposure variables to be risk factors for human infestation: low degree of education of household leader (OR=3.9 [95% CI 2.3–6.7]), house built of palm stems (OR=4 [95% CI 1.9–8.6]), kitchen stall on compound (OR=1.6 [95% CI 1.1–2.5]), and male sex (OR=2.2 [95% CI 1.5–3.2]; all p<0.02). The mere presence of a pet animals in a household was no risk factor for human infestation with *T. penetrans*. In contrast, however, the presence of infested animals was (OR=1.6 [95% CI 1.1–2.3], p=0.01). In the logistic regression, sex, education level, type of housing, and the presence of infested animals remained as independent risk factors for human tungiasis, irrespective of whether the pet animal was a dog or a cat (Table 3).

In individuals living together with at least one infested pet animal, the intensity of infestation was significantly higher (mean=3.4 [95% CI 2.3–4.6]) than in household members without pet animals or only with healthy animals in their household (mean=0.6 [95% CI 0.3–1]; p=0.01). The intensity of infestation in humans correlated with the

 Table 2
 Animal species present in Balbino village and proportion infested with *T. penetrans*

Species	Animals examined N	Animals infested (%; 95% CI)
Dogs	61	(68.9; 55.7-80.1)
Cats	63	(49.2; 36.4-62.1)
Horses	11	0
Donkeys	8	0
Oxen	5	0
Pigs	5	0

CI Confidence interval

Exposure variable	Odds ratio	95% confidence interval	<i>P</i> -value
Male sex	2.64	1.73-4.03	< 0.01
Low education level ^a	2.76	1.58-4.84	< 0.01
House built of palm stems	3.78	1.71-8.35	< 0.01
Infested animal present in	1.69	1.1–2.6	0.017
household			

 Table 3 Multivariate regression analysis of risk factors associated with the presence of tungiasis in humans

^a Only primary school completed by household leader

intensity of infestation in animals (rho=0.19, p=0.01; Fig. 1).

The relationship between intensity of infestation in animals and humans was even more pronounced at the household level. In households with infested animals, the median number of lesions was six (interquartile range 0–25) compared to two (interquartile range 0–6) in households without infested animals (p=0.01). In households with infested animals, the percentage of infested household members was higher than in households without infested animals: 27% (95% CI 20–33%) versus 42% (95% CI 30–53%), p=0.02. The percentage of infested individuals per household correlated with the number of infested animals (rho=0.3, p=0.02).

Discussion

Tungiasis is a common zoonotic skin disease in many resource-poor communities of Latin America, the Caribbean, and sub-Saharan Africa (Ade-Serrano and Ejezie 1981;

Fig. 1 Correlation between intensity of *T. pentrans* infestation in humans and animals living in one household (linear regression line; rho=0.19; p=0.01) Chadee 1998; Heukelbach et al. 2001). Depending on the endemic area, *T. penetrans* parasitizes a broad range of sylvatic and domestic animals such as monkeys, elephants, armadillos, sheep, cattle (Franco da Silva et al. 2001), goats (Trentini 2000), pigs (Njeumi et al. 2002; Pampiglione et al. 1998; Ugbomoiko et al. 2007b), rats, cats, and dogs (Heukelbach et al. 2004; de Carvalho et al. 2003).

During the last years, evidence has accumulated pointing towards the importance of animal reservoirs for the occurrence of tungiasis in human population (de Carvalho et al. 2003; Heukelbach et al. 2004; Muehlen et al. 2006; Njeumi et al. 2002; Pampiglione et al. 1998; Ugbomoiko et al. 2007a). However, it remains uncertain to what extent animals contribute to human infestation with T. penetrans in different settings. This is a matter of concern as factors that determine high prevalence and intensity of infestation in the human population also contribute to the presence of severe morbidity (Kehr et al. 2007). Moreover, control measures will be more effective if the role of reservoirs and transmission dynamics are better understood. Therefore, tungiasis in the human and the animal population of an endemic village was scrutinized and the presence of the ectoparasitosis and the intensity of infestation were correlated in both groups.

According to Sampaio (1976) and Linardi (2000) pig are an important reservoir for *T. penetrans* in rural areas of Brazil. In Africa, pigs are the most important reservoir for this parasite, (Pampiglione et al. 1998; Ugbomoiko et al. 2007b) causing considerable losses in piglet farms (Verhulst 1976). However, in the present study, pigs were not found infested. While dogs and cats could wander around in the village, pigs were kept in little shacks on the compound. The containment was a consequence of a law-enforced health policy intending to reduce cysticercosis. Thus, it seems that



Median number of sand fleas per household in dogs and cats

the preventive measure targeted against cysticercosis also contributed to the absence of tungiasis in pigs. In the shacks, the sandy soil remained wet with feces and urine. The subsequent change in soil acidity presumably makes the development of sand flea larva impossible (Newstead 1909).

In the nineteenth century, travelers who had used abandoned huts for overnight stays in the endemic area became heavily infested with T. penetrans (Bonnet 1867; Karsten 1865). In and around these houses, rats and mice had been frequent and presumably infested with sand fleas (Bonnet 1867; Karsten 1865). These anecdotal observations point to an ongoing cycle in the absence of humans in substandard houses and or to the presence of off-host stages remaining vital in the soil for months and able to hatch in the presence of an appropriate stimulus, e.g., arrival of a human being (Heukelbach et al. 2004). Assumably, rats could act as the principal reservoir in this situation, but other rodent species could maintain the cycle as well (Ugbomoiko et al. 2007b). Using laboratory-raised rats, Witt et al. (2007) showed that they became rapidly infested in poor urban neighborhoods when placed indoors or outdoors at preferred whereabouts of dogs and cats (Witt et al. 2007).

The fishing community Balbino differs from the urban slum in as much as R. rattus was not found and other sylvatic rodents are rarely seen (Heukelbach et al. 2004). Still, it is conceivable that animals with intimate contact to humans such as dogs and cats carry T. penetrans into or near to the house. In a risk factor study in a rural settlement in Brazil, Muehlen et al. (2006) found the presence of tungiasis in humans associated with dogs living in the household. As dogs are an important reservoir for tungiasis in Brazil (Heukelbach et al. 2004), the authors concluded that peri- and intra-domiciliary transmission of T. penetrans might be enhanced by dogs; although due to the study design, they could not provide further evidence for this hypothesis (Muehlen et al. 2006). However, data from Africa support this assumption that the presence of animals in or near to the house increases the risk of tungiasis in household members. Ugbomoiko et al. (2007b) identified pigs as the most important predictor for the presence of tungiasis in inhabitants in a Nigerian village, if the animals were kept on the compound (Ugbomoiko et al. 2007b). The present study confirms the previous observations by showing that dogs and cats contribute to a high frequency of tungiasis and a high intensity of infestation in the household.

The observation that on the household level the intensity of infestation in pet animals and humans correlates emphasizes animal–human interactions in the transmission of *T. penetrans*. The female sand flea expels dozens of eggs per day which may hatch indoors and outdoors, if conditions are favorable (Linardi 2000). In Balbino village, dogs and cats have intimate contact with humans and rest near to or in the house. When infested with *T. penetrans*, their presence will augment

the number of adult sand fleas intra- and peri-domiciliary, which is expected to result in high attack rates in humans.

The present study has two limitations. Due to its design as a cross-sectional study, it can only be assumed that a cause–effect relation exists between animal and human tungiasis. Furthermore, it remains uncertain whether infested cats and dogs contribute predominantly to a perior to an intra-domiciliary transmission because the precise whereabouts of these animals were not determined.

The zoonotic character of tungiasis makes control in affected communities more complex than for other ectoparasitic diseases without an animal reservoir, such as scabies or head lice (Heukelbach et al. 2003). The widespread occurrence of domestic animals and rodents in resource-poor communities and the high prevalences of tungiasis in these animals make it clear that veterinary aspects have to be integrated in the control of this zoonosis. Klimpel et al. (2005) showed that the regular treatment of animals with the combination of a dermal solution containing imidacloprid and permethrin lowered the parasite burden in the treated animals. So far, clinical observations exist indicating that integrated control measures could lead to a decline in the incidence of tungiasis in humans (Heukelbach et al. unpublished observation).

In conclusion, the data obtained here identified parasitized dogs and cats, man's closest companions, as risk factors for tungiasis in the household. Their presence gives an important ad on to an intensity of infestation and therefore, on the population level, contributes to severe morbidity in humans. Control measures designed to reduce the prevalence and intensity of infestation in humans must therefore include especially companion animals such as dogs and cats and their regular treatment using ectoparasiticides.

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