EFFECT OF *EL NIÑO* PHENOMENON ON THE SEASONALITY OF *Pulicidae* (*Siphonaptera*) IN DOGS IN A RURAL AREA OF SOUTHERN BRAZIL

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ABSTRACT

he aim of this study was to investigate the effect of *El Niño* phenomenon on the seasonality of flea species present on dogs from a rural area in Southern Brazil, and on the relationship between the frequency of these species and the climatic conditions in that micro-region. Over three years (2013-2016) 8,974 fleas were collected from 20 dogs. *Ctenocephalides felis* was the most frequent species (70.14%), followed by *Pulex irritans* (28.82%) and *Ctenocephalides canis* (1.05%). The number of C. felis and *C. canis* individuals varied significantly with the year and with the season of the year, whereas the number of *P. irritans* was only influenced by the season. C. felis was more prevalent than all the other species in all seasons studied. There was a significant difference in the number of specimens collected during 2015, under the influence of *El Niño*, compared to 2013 but not to 2014. There was a higher incidence of *P. irritans* during the cooler periods. *C. canis* population increased during colder seasons whereas no individuals were collected in years with lower average temperatures. The present study expanded the current knowledge about the seasonal variations in the *Siphonaptera* populations parasiting dogs, especially in the years under influence of *El Niño*.

Keywords: Fleas. Ctenocephalides felis. Ctenocephalides canis. Pulex irritans.

INTRODUCTION

Fleas are wingless insects with laterally flattened body, bloodsucking, holometabolous parasites of mammals and birds (DOBLER; PFEFFER, 2011). They evolved between the Cretaceous and the Jurassic, 125 million to 150 million years ago, parasitizing initially marsupials and other small mammals (DURDEN; TRAUB, 2009).

Ctenocephalides felis Bouché 1835 is the most frequent species found in dogs and cats in Europe and in America (CASTRO; RAFAEL, 2006; STALLIVIERI et al., 2009), while Ctenocephalides canis Curtis 1826 seems to have a more restricted geographical distribution (MARCHIONDO et al., 2007), better adapting to low temperature regions (GUIMARÃES et al., 2011; OLIVEIRA; RIBEIRO, 1983). Pulex irritans Linnaeus 1758 is a cosmopolitan Pulicidae of eclectic food preferences whose adults usually do not reside on the body of the host only seeking the host for biting (LINARDI, 2011). There are reports from several countries showing that this flea parasitizes different species of domestic animals such as dogs and cats (DOBLER; PFEFFER; 2011), sheep, goats, cattle and pigs (DOMÍNGUEZ-PEÑAFIEL et al., 2011; KAAL et al., 2006; RAHBARI et al., 2008).

In addition to the blood loss from flea biting, 50% of dermatologic cases seen in veterinary clinics in dogs and cats are believed to be due to flea biting (BITAM et al., 2010; TRAVERSA, 2013). As pathogen transmitters, *C. felis* and *P. irritans* are among the vectors of Rickettsia tiphy, the causative agent of Murine Typhus, for which there are already cases of infected humans reported in Brazil. *C. felis* transmits Bartonella henselae and B. clarridgeiae, the causative agents of Cat-scratch Disease (PERSICHETTI et al., 2016; TRAVERSA, 2013). *P. irritans* plays an important role in the transmission among human hosts of Yersinia pestis, the causative agent of bubonic plague (BITAM et al., 2010). Recently *Rickettsia felis* was identified in febrile patients in sub-Saharan Africa and *C. felis* is the only confirmed vector of this pathogen, although transmission by Anopheles mosquitoes has not been ruled out (ANGELAKIS et al., 2016).

The *El Niño* Southern Oscillation (ENSO) is a large-scale atmospheric-oceanic phenomenon characterized by temperature abnormalities on the surface water of the Equatorial Pacific Ocean, which causes heat and cold extremes in the Southern cone of South America, and

large interannual variations of rainfall. In the negative phase of the phenomenon, called La Niña, rainfall is generally below normal levels, while in its positive phase, *El Niño*, there is an increase of average winter temperatures and higher rainfall during late winter and the entire spring (GRIMM et al., 2000).

As for other insects, the diversity and abundance of fleas varies significantly among geographic regions and micro-regions as well, according to climatic variations and seasonality (COSTA-JUNIOR et al., 2012). Insect population fluctuations have been associated with climate change, particularly to *El Niño* (DEJEAN et al., 2011). The increase in cases of bubonic plague due to the increase of rainfall related to *El Niño*, with a consequent increase in the populations of rodents and their fleas, was documented by Parmenter et al. (1999) in New Mexico (USA), Dávalos et al. (2001) in Peru, and Moore et al. (2015) in Uganda.

Previous research suggested that climate variability has a direct influence on the epidemiology of vector-borne diseases (GAGE et al., 2008). Indeed, abundance of fleas is affected by climate dynamics such as environment temperature, rainfall rates, and relative humidity, with warm-moist weather providing a likely explanation for higher flea frequencies (DURDEN; TRAUB, 2009; KRASNOV, 2008). Thus, gathering information on the effects of climatic factors affecting the populations of *Siphonaptera* dog parasites may be the key to the planning of integrated ectoparasite control programs. Indeed, an understanding of the relationship between the climate dynamics of *El Niño* and the populations of different flea species could be useful in predicting periods of increased infestation and risk of pathogen transmission by these insects, requiring stricter control measures.

The aim of this study was to investigate the effect of *El Niño* on the seasonality of species of fleas sampled from dogs from a rural area of Southern Brazil and the relationship between the frequency of these species and the climatic conditions of temperature and rainfall rates in this micro-region.

MATERIAL AND METHODS

For three years (February 2013 to January 2016), 10 to 15 fleas were sampled monthly from each of 20 dogs, naturally infected by fleas, from a rural property in the municipality of Bagé, in the state of Rio Grande do Sul (RS), Brazil, located at 30°55'53.53" S, 53°36'51.28" W, at an

elevation of 145 meters. The property is a farm that breeds beef cattle, sheep, goats, pigs and poultry, where dogs are used for work and company, as both herding and hunting dogs. No regular parasite control measures, for either ecto or endoparasites, were used in these dogs. Monthly climatic data of the region were obtained from the National Institute of Meteorology of Brazil (INMET, 2016).

Fleas were captured by application of about 100 ml ethanol 92.6% ABV per animal, directly to the skin along the spine. Fleas that jumped out, due to the repulsive effect of alcohol, were collected with histological tweezers and placed in plastic bottles containing alcohol 70% ABV, labeled with a sample number, and later identified by species and sex at the Parasitology Laboratory of Universidade da Região da Campanha, Bagé, RS, Brazil, through a stereomicroscope, with the help of identification keys (BICHO; RIBEIRO, 1998; LINARDI; SANTOS, 2012).

The number of fleas was first normalized by the square root of X + 1. Then, the number of fleas of each species was evaluated by two-factor analysis of variance (ANOVA), and the year and season when they were collected were added as independent variables (p=0.05). The mean number of fleas from each of the species found in the seasons of the year were compared by Tukey test (p=0.05). All analyzes were performed with SPSS, version 22.0 for Windows (IBM, ARMONK, NY, USA, 2013).

This work was approved by the Ethics Committee on Animal Experimentation of the Universidade Federal de Pelotas, Rio Grande do Sul, Brazil, under the Process 23110.004046/2013-89.

RESULTS AND DISCUSSION

All 20 dogs studied herein were infested with fleas during the entire trial period (three years), and a total of 8,974 specimens were collected. *C. felis* was the most common species (6,294; 70.14%), followed by *P. irritans* (2,586; 28.82%) and *C. canis* (94; 1.05%) (Table 1).

For *P. irritans*, the female: male ratio was roughly balanced (1.03:1), whereas, for *C. felis* and for *C. canis* the proportion exceeded two females for each male (2.84:1 for *C. felis* and 2.48:1 for *C. canis*).

Table 1 - Siphonaptera species collected in dogs, relative frequency and sex ratio, annual and total in a farm in the Southern Brazil. From February 2013 to January 2016.

Flea species	N₀ (Q, ठ)	Relative frequency (%)	Sex ratio
Ctenocephalides felis TOTAL	6,294 (1,640♂ 4,654♀)	70.14	0.74 (1: 2.84)
YEAR 1	1,907 (505♂ 1,402♀)	63.78	0.73 (1: 2.78)
YEAR 2	2,202 (558♂ 1,644♀)	72.15	0.75 (1: 2.95)
YEAR 3	2,158 (577♂ 1,608♀)	74.29	0.74 (1: 2.79)
Ctenocephalides canis TOTAL	94 (27♂ 67♀)	1.05	0.71 (1: 2.48)
YEAR 1	86 (25♂ 61♀)	2.88	0.71 (1: 2.44)
YEAR 2	8 (2♂ 6♀)	0.26	0.75 (1: 3.00)
YEAR 3	0	0	0
Pulex irritans TOTAL	2,586 (1,276♂ 1,310♀)	28.82	0.51 (1: 1.03)
YEAR 1	997 (482♂ 515 ²)	33.34	0.52 (1: 1.07)
YEAR 2	842 (406♂ 436♀)	27.59	0.52 (1: 1.07)
YEAR 3	747 (388♂ 359♀)	25.71	0.48 (1: 0.92)
TOTAL	8,974 (2,943♂ 6,031약)	100.00	0.67 (1: 2.05)

The number of C. felis individuals was influenced both by the year and by the seasons (F = 4.268, DF = 2, p<0.05, and F = 8.125, DF = 3, p<0.001, respectively), and the same was observed for *C. canis* (F = 54.341, DF = 2; p<0.001, and F = 2.996, DF = 3, p=0.05, respectively, for years and seasons). However, the number of P. irritans was not influenced by the year (F = 2.665, GL = 2, p=0.090), but was influenced by the seasons (F = 4.935, GL = 3, p<0.05). The seasonal frequency distribution (summer, fall, winter and spring) of the species of fleas is shown in Figure 1. The average monthly temperature (°C), rainfall (mm) rate and relative humidity (% RH) for the three years are displayed in Figure 2.

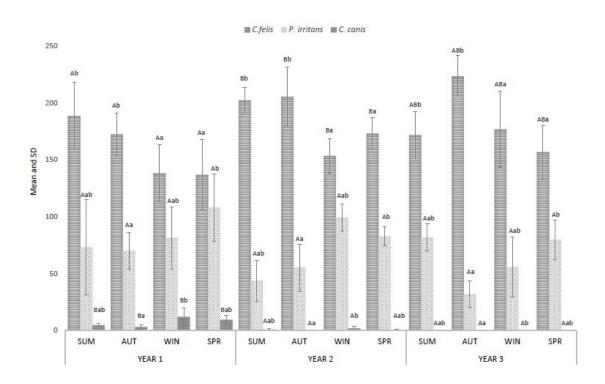


Figure 1 - Seasonality of *Ctenocephalides felis, Ctenocephalides canis* and *Pulex irritans*, from February 2013 to January 2016, on dogs from a farm in the Southern Brazil. Standard deviations are included. Different capital letters show significant differences in the mean comparison between years and lower case letters show significant differences in the mean comparison between seasons, by Tukey test (p=0.05).

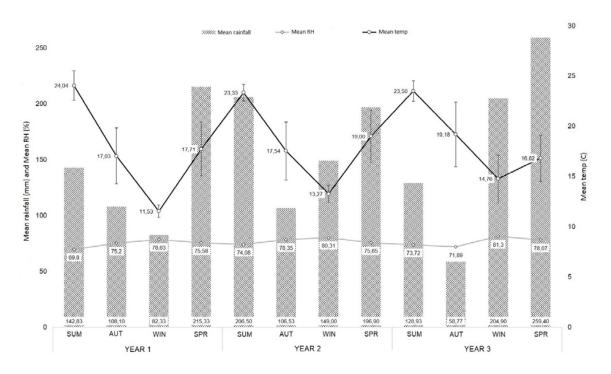


Figure 2 - Seasonal average temperature (°C), rainfall (mm) and relative humidity (%) from February 2013 to January 2016, in Bagé, RS, Brazil (INMET, 2016).

During the winter of 2014, average temperature was 1.74 oC higher than 2013, and in 2015 average temperature was 1.49 oC higher than 2014, and 3.23 oC higher than 2013. In addition, the temperature in the fall of 2015 was 1.64 oC higher than that of the same season in the previous year.

Ctenocephalides felis was more prevalent than all other species for all seasons, during the three years of the study. However, there was a significant variation in the number of specimens collected during 2015, which was under the influence of *El Niño*, if compared to the first year of the experiment (Figures 1, 2 and 3). No difference was observed between 2015 and the previous year (2014), even though, in 2014, average winter temperatures were higher than those in 2013.

The time series presented in Figure 3 shows the increase of *C. felis* specimens during the fall months, with greater abundance during the fall of 2015. In contrast, the number of collected specimens of *P. irritans* shows that this species tends to be in greater abundance in colder times of the year, with an inverse relation to *C. felis*.

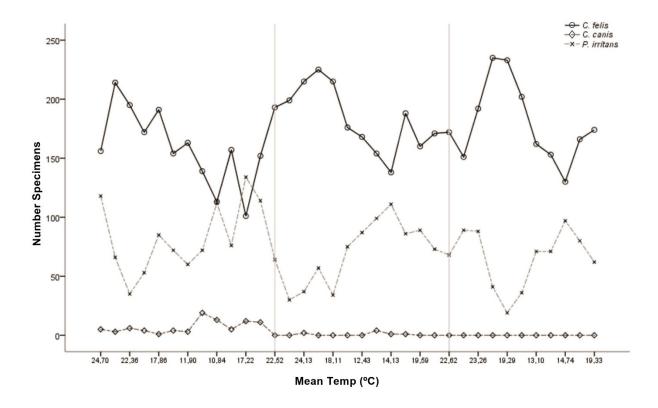


Figure 3 - Time series of the number of *Ctenocephalides felis* specimens, *Ctenocephalides canis* and *Pulex irritans*, according to the average temperature from February 2013 to January 2016, in dogs from a farm in the municipality of Bagé, RS, Brazil.

Ctenocephalides canis population clearly increased in colder seasons and in years with a lower average temperature (Figures 1, 2 and 3). In the year 2015, with the occurrence of the El Niño phenomenon and the consequent increase in average temperatures in winter by 3.23 oC compared to the first year (2013), no individuals of this species were collected.

Regarding the sex ratio, the data found in our study (Table 1) are consistent with results reported by Linardi e Nagem (1973) for the species *Pulex irritans* (1.03:1) and Oliveira e Ribeiro (1983) and Rodrigues et al. (2001) for *C. felis* and *C. canis* (2.84:1 and 2.48:1), respectively.

The trend of increase in autumn-winter temperatures is characteristic of the *El Niño* phenomenon (DEJEAN et al., 2011), but, in 2015, Southern Brazil presented the highest temperatures ever recorded and a significant increase in rainfall in the winter and spring (TELLEFSON, 2016). Accordingly, the present study shows that the average rainfall in the

winter, and especially in the spring of the third year of the experiment (2015), was much higher than in the previous two years.

Ctenocephalides felis is the flea species most prevalent in dogs in Brazil and in tropical and temperate regions around the globe (COSTA-JUNIOR et al., 2012; DRYDEN; RUST, 1994). This prevalence was confirmed herein, since we found much higher prevalence of this species than the others throughout the whole period studied, with significant increase in infestations during the fall months.

In this study, the relative frequency of *P. irritans* was 28.82%. In Greece, a frequency of 0.8% was described (KOUTINAS et al., 1995), in Chile 18.8% (ALCAÍNO et al., 2002) and, in a Spanish resort, up to 100% were found (GRACIA et al., 2000). According to Rust et al. (1971), when various hosts are available in the same niche, *P. irritans* prefers dogs. Also, *P. irritans* can more easily infect humans than *C. felis* or *C. canis* (HARMAN et al., 1987).

The higher incidence of *P. irritans* during colder times of the year (Figures 1-3) is noteworthy, as this species has a Neotropic origin (LEWIS, 1972). The Pulicidae may be changing its habits of seeking the host only for feeding (LINARDI, 2011), as the canine host also provides protection against abiotic conditions. In another study (SILVA et al., 2016), our research group, has shown the preference of *P. irritans* for long-haired dogs, confirming a trend for this flea to remain in the dogs, thus changing their eating behavior. Furthermore, the increase of *P. irritans* population during periods when the population of *C. felis* decreased may indicate that *P. irritans* is host-opportunistic and occupies a niche left by the host-specific *C. felis*, when the latter species has its population smaller due to unfavorable abiotic conditions.

Several authors have suggested that *C. canis*, of Palearctic origin, prefers cooler climates (BECK et al., 2006; GONZÁLEZ et al., 2004; RODRIGUES et al., 2001). This observation is confirmed herein, since in the autumn-winter of 2015, when the temperatures rose well above average, no *Siphonaptera* of this species were collected.

Understanding the relationship between climate and *Siphonaptera* parasite populations on dogs is of utmost importance in the planning of integrated ectoparasite control programs,

because the seasonal distribution of the species can be informative as to when major infestations are likely to occur.

CONCLUSION

We show that infestations are more significant in *El Niño* years. The number of *C. felis* and *C. canis* individuals varied significantly with the year and with the season of the year, whereas the number of *P. irritans* was only influenced by the season.

EFEITO DO FENÔMENO *EL NIÑO* SOBRE A SAZONALIDADE DE PULICÍDEOS (*Siphonaptera*) EM CÃES DA ZONA RURAL DO MUNICÍPIO DE BAGÉ, RS, BRASIL

RESUMO

objetivo deste trabalho foi verificar o efeito do fenômeno El Niño sobre a sazonalidade das espécies de pulgas em cães de uma zona rural do Rio Grande do Sul, Brasil, e a relação da freguência dessas com as condições climáticas nessa microrregião. Durante três anos (2013-2016), foram coletadas 8.974 pulgas em 20 cães. Ctenocephalides felis foi a espécie mais frequente (70,14%), seguida por Pulex irritans (28,82%) e Ctenocephalides canis (1,05%). O número de indivíduos de C. felis e C. canis foi influenciado significativamente pelo ano e pelas estações do ano, enquanto que o número de P. irritans foi influenciado apenas pelas estações do ano. C. felis alcançou maior prevalência do que as demais espécies em todas as estações, durante os três anos do estudo. Houve variação significativa entre o número de espécimes coletados durante o último ano (2015), sob influência do El Niño, em relação ao primeiro ano do experimento, mas não em relação ao ano anterior (2014). Houve maior ocorrência de P. irritans durante épocas mais frias em todos os anos do experimento. C. canis teve aumento populacional em épocas mais frias e em anos com temperatura média mais baixa. No ano de 2015, com a ocorrência do fenômeno El Niño e consequente aumento das temperaturas médias invernais não foram coletados indivíduos dessa espécie. O presente estudo permitiu aprofundar o conhecimento atual sobre as variações sazonais das populações de sifonápteros parasitos dos cães, principalmente nos anos de ocorrência do El Niño.

Palavras-chave: Pulgas. Ctenocephalides felis. Ctenocephalides canis. Pulex irritans.

EFECTO DEL FENÓMENO EL NIÑO SOBRE LA DISTRIBUCIÓN ESTACIONAL DE PULÍCIDOS (Siphonaptera) EN PERROS DE LA ZONA RURAL DEL MUNICIPIO DE BAGÉ, RS, BRASIL

RESUMEN

■ l objetivo del trabajo fue avaluar el efecto del fenómeno El Niño sobre la distribución estacional de pulgas en perros de una región rural del Rio Grande do Sul, Brasil, y la relación de frecuencia de esas especies con las condiciones climáticas de temperatura y precipitación pluviométrica en esa microrregión. Durante tres años (2013-2016), fueron colectadas 8.974 pulgas en 20 perros. Ctenocephalides felis fue el espécimen más frecuente (70,14%), seguida por Pulex irritans (28,82%) y Ctenocephalides canis (1,05%). El número de individuos de C. felis e C. canis fue influenciado significativamente por el año y por las estaciones del año, en cuanto el número de P. irritans fue influenciado apenas por las estaciones del año. C. felis tuvo mayor prevalencia que las otras especies en todas estaciones, durante los tres años. Hubo variación significativa entre el número de especímenes colectados durante el último año (2015), con influencia de El Niño, en relación al primer año, pero no en relación al año anterior (2014). Hubo mayor ocurrencia de P. irritans durante épocas más frías del año, en todos los años del trabajo. C. canis tuvo aumento de población en épocas más frías y en años con temperatura media más baja. En el año 2015, con la ocurrencia de El Niño y el aumento de las temperaturas medias invernales, no fueron colectados individuos de esa especie. El presente estudio permitió ahondar el conocimiento actual sobre las variaciones estacionales de poblaciones de sifonápteros parásitos de perros, principalmente en años de ocurrencia de El Niño.

Palabras clave: Pulgas. Ctenocephalides felis. Ctenocephalides canis. Pulex irritans.

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