# Age structure of owned dogs under compulsory culling in a visceral leishmaniasis endemic area

A estrutura etária de cães com donos numa área endêmica de leishmaniose visceral

Estructura etaria de la población canina domiciliada en un área endémica con leishmaniasis visceral canina

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#### **Abstract**

The age structure of the dog population is essential for planning and evaluating control programs for zoonotic diseases. We analyzed data of an owned-dog census in order to characterize, for the first time, the structure of a dog population under compulsory culling in a visceral leishmaniasis endemic area (Panorama, São Paulo State, Brazil) that recorded a dog-culling rate of 28% in the year of the study. Data on 1,329 households and 1,671 owned dogs revealed an owned dog:human ratio of 1:7. The mean age of dogs was estimated at 1.73 years; the age pyramid indicated high birth and mortality rates at the first year of age with an estimated cumulative mortality of 78% at the third year of age and expected life span of 2.75 years. In spite of the high mortality, a growth projection simulation suggested that the population has potential to grow in a logarithmic scale over the years. The estimated parameters can be further applied in models to maximize the impact and minimize financial inputs of visceral leishmaniasis control measures.

Dogs; Age Distribution; Visceral Leishmaniasis; Zoonoses

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#### Introduction

Availability of data on the demographic structure of dog populations is essential for the successful planning and evaluation of programs designed to control zoonotic diseases 1 such as rabies and visceral leishmaniasis (VL). This vector-borne tropical disease is caused by Leishmania infantum (also known as Leishmania chagasi) and has the domestic dog (Canis lupus familiaris) as its main urban reservoir 2. The disease is widespread in South America 3, and Brazil, along with another five Latin American countries, make this region responsible for 90% of global human cases of VL 4. Currently, the Brazilian Ministry of Health recommends vector elimination by residual insecticide spraying and identification and culling of positive dogs as control measures for VL, as well as the early diagnosis and treatment of human cases. Additional control measures such as birth control by mass sterilization have been proposed as a public health policy to replace euthanasia.

While a large body of literature has addressed the controversy as to whether or not the elimination of positive dogs is associated with a reduction in VL incidence in humans 5,6,7,8,9, the impact of increased mortality in the structure and composition of canine populations has not been assessed in spite of its potential to influence VL prevalence in dogs and, consequently, in humans 7,10. Furthermore, the replacement after compulsory culling also affects the structure of the canine population, thus changing the impact of currently used VL control measures.

Here, we analyzed data of a canine census in order to characterize, for the first time, the structure of a dog population under compulsory culling in a VL endemic area. The estimated parameters may help the evaluation of proposed and currently used control measures in Brazil.

## Material and methods

## Ethical statement

This study was carried out in strict accordance with the recommendations in the Ethical Principles of the Brazilian College of Animal Experimentation (COBEA; http://www.cobea.org.br). The protocol was approved by the UNESP Ethics Committee on Animal Experimentation (process 2010/003389).

#### Study area

The study was conducted in the municipality of Panorama, located in the Northwest region of the state of São Paulo (latitude 21º21'23" S, longitude 51°51'35" W). The municipality has an estimated population of 15,374 inhabitants (http://www. ibge.gov.br/cidadesat/, accessed on 03/Jul/2015) and no demographic data about the population of owned dogs has been published.

Since 2007, the municipality has been endemic for canine visceral leishmaniasis (CVL) and the actions employed to control CVL in the area follow the Brazilian Ministry of Health recommendations: health education, early diagnosis and treatment of human cases, vector control by residual insecticide spraying in the residencies around the area of a positive human case, and serological screening with subsequent compulsory culling of the VL positive dog 11.

CVL seropositivity ranged from 29.5% (2007) to 41.9% (2010) and started decreasing thereafter (30.6% in 2011; 20.2% in 2012). Compulsory culling was conducted by the municipality's Center for Zoonosis Control (CZC) in rates ranging from 20.4% (576/2,825) in 2007 to 28.2% (407/1,444) in 2010; thereafter culling decreased to 16.1% (80/496) in 2011 and 3.75% (53/1,410) in 2012. Of note is the lack of diagnostic kits in the years of 2011 and 2012 which bias the decrease of the euthanasia rates. Before the approval in 2008 of a state law that regulated euthanasia, unwanted dogs were sent to culling by their owners for reasons other than VL (moving, aggression, costs) representing 11 to 27% of the population (unpublished data obtained from CZC records).

VL human incidence ranged from 0.82/1,000 inhabitants (2007) to 0.068/1,000 inhabitants (2012) 12.

## Descriptive study

The data analyzed here were obtained from a household census conducted between February and March 2010 by applying a door-to-door questionnaire. The questionnaire was filled out by health agents who were used to handle dogs once they had sampled them for the routine seroepidemiological CVL survey. They collected data on the number of animals per household, gender, age (informed by the owner and checked by the agents), breed, and number of viable offspring per female. Information regarding this latter variable was also given by the owner, once the health agent had made it clear that "offspring per female" meant the number of puppies that were born and survived.

It is noteworthy that health agents were trained to apply the questionnaire in order to maximize the accuracy of the data. Due to the fact that this study was based on age information, although examination of the teeth was not possible, health agents were trained by one of the veterinarian authors (D.V.B.) to check whether the dogs appeared to be the age that had been informed, especially in the cases of senile (> 7 years old) and young dogs (< 2 years old). Although some degree of imprecision was expected in the recorded ages of adult animals, the age assignment in young and senile dogs was deemed to be fairly accurate. Houses were revisited at least once if they were initially closed.

The dataset was analyzed using methods summarized by Pianka 13, using locally developed scripts written in R version 3.0.1 (The R Foundation for Statistical Computing, Vienna, Austria; http://www.r-project.org).

#### Age structure

In order to descriptively assess the canine age distribution in Panorama, a static (vertical) life table and a population pyramid were constructed in R version 3.0.1. First, animals were sorted into w = 8 age-classes of width equal to 1 year, starting from zero and ending at dogs over 7 years of age. Each class included its lower boundary value in the calculation of the grouped frequency. Based on the absolute frequency of individuals per age-class x, represented by n(x), the following components of the static life table were calculated:

Cumulative survival = l(x). Proportion of individuals still alive at age-class x relative to the first age-class (x = 1):

$$l(x) = \frac{n(x)}{n(x=1)} \tag{1}$$

Age-class specific survival = p(x). Proportion of survivors from age-class x to age-class x + 1:

$$p(x) = \frac{n(x+1)}{n(x)} \tag{2}$$

Cumulative mortality = d(x). Proportion of extinct individuals at age-class x relative to the first age-class (x = 1):

$$d(x) = 1 - l(x) \tag{3}$$

Age-class specific mortality = q(x). Proportion of extinct individuals from age-class x to age-class x + 1:

$$q(x) = 1 - p(x) \tag{4}$$

Age-class specific life expectancy = e(x). Expected number of time units (in this case, in years) an individual in age-class x will live:

$$e(x) = \frac{\sum_{y=x}^{w} l(y)}{l(x)}$$
 (5)

#### Growth projection

Additional statistics taking only female data into account were calculated to assess population growth. First, the age-class-specific average *number of viable offspring per female* = m(x) was calculated as follows: Let np(x) represent the number of puppies in age-class x and nf(x) the number of females in age-class x. The number of puppies born per female and age-class was calculated as:

$$m(x) = \frac{n p(x)}{n f(x)} \tag{6}$$

As only viable offspring were considered in the calculation, the age-class specific realized fecundity = F(x) was considered interchangeable with m(x). Given the present date age distribution, the proportions of survivors from each age-class to the next, and the age-class specific fecundities, the age-classified Leslie matrix model 14,15 was adopted in order to project the dog population growth, assuming a constant environment and a population with fixed survival and fecundity. Thus, the transition matrix A was constructed based on the estimated values of F(x) and p(x):

$$A = \begin{pmatrix} F(1) & F(2) & F(3) & F(4) & F(5) & F(6) & F(7) & F(8) \\ p(1) & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & p(2) & 0 & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & p(3) & 0 & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & p(4) & 0 & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & p(5) & 0 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & p(6) & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 & 0 & p(7) & 0 \end{pmatrix}$$
 (7)

Starting with the vector of absolute frequencies of individuals per age-class n(x), the age frequency distributions were iteratively estimated, for each time t, as:

$$n^t = A n_{t-1} \tag{8}$$

Age distributions were estimated for 20 iterations. Population growth was projected as the simulated total number of individuals for each iteration over time. The asymptotic finite rate of growth of the population ( $\lambda$ ) was obtained as the dominant eigenvalue of a singular value decomposition of matrix A. The intrinsic rate of population growth was calculated as  $r = ln(\lambda)$ . The stable age distribution was obtained as the right eigenvector of the transition matrix (i.e., column

eigenvector of  $\lambda$ ). Age-class specific reproductive values, v(x), representing the expected number of future offspring per female in each age-class, were obtained as the first row vector of the inverted matrix of right eigenvectors. Reproductive values were scaled by dividing each v(x) by the reproductive value of age-class x=1. Finally, two additional fecundity parameters were estimated: Net reproductive rate =  $R_0$ . Average number of offspring per female per generation (i.e. replacement rate per female):

$$R_0 = \sum_{x=1}^{w} l(x) m(x)$$
 (9)

*Generation time* = *T*. Number of time units (in this case, in years) it takes a female to produce one viable offspring:

$$T = \frac{\sum_{x=1}^{w} x l(x) m(x)}{R_0} \tag{10}$$

#### Results

Figure 1

A total of 6,271 households were visited in the urban area of Panorama, among which 1,329 (21.2%) owned a total of 1,950 dogs, resulting in 0.31 dogs per household. Considering only

households where dogs were present, the average number of dogs per household was approximately 1; the owned dog:human ratio was 1:7. Replacement reported by the owners during the door-to-door visits was 7.8% (93 out of 1,194).

After filtering the dataset for missing values (observations for which the data table was not completely filled), 1,671 (85.7%) observations remained, and the male:female ratio was estimated at approximately 1:1. Ages ranged from 0 to 17 years, and the mean age was found to be 1.73 years, with a 95% confidence interval of 1.62-1.84. The age pyramid (Figure 1) followed an expansive pattern (i.e., it was very wide at the base and very narrow at the top).

The age-structured data was used to build a vertical life table (Table 1). The first age class (from birth to 1 year of age) accounted for 50.7% of the contingency. The highest proportion of survivors and lowest mortality was found in age-class 6 to 7 years, with virtually all dogs transitioning to the subsequent class (p(x) = 1 and q(x) = 0). The first age-class exhibited the lowest proportion of survivors and the highest mortality among all classes (p(x) = 0.33 and q(x) = 0.67).

The rapid decay in survivorship suggested that the greatest mortality happens early in life in this population, and is particularly high in the first 12 months (Figure 2). The expected life span

Gender-specific age pyramids for the studied canine population.

## February-March/2010

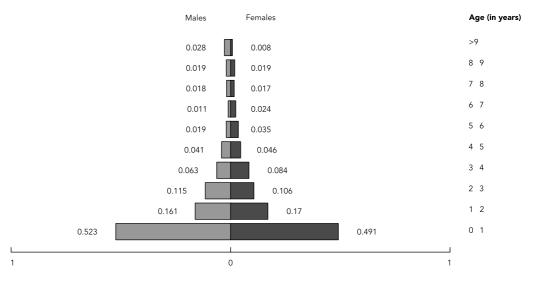


Table 1

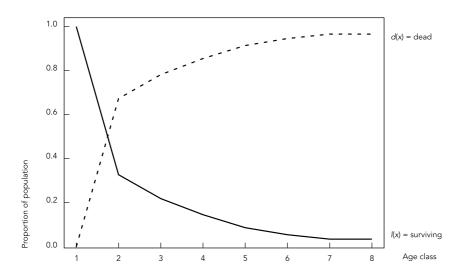
Vertical life table for the canine population in the study.

Age class	n(x)	%n(x)	Stable age	l(x)	p(x)	d(x)	q(x)	e(x)	m(x)
			distribution						
0  -1	847	50.69	56.41	1.00	0.33	0.00	0.67	1.90	0.31
1 -2	277	16.58	17.73	0.33	0.67	0.67	0.33	2.75	0.57
2  -3	185	11.07	10.06	0.22	0.66	0.78	0.34	2.62	1.36
3  -4	123	7.36	7.21	0.15	0.59	0.85	0.41	2.44	1.10
4   5	73	4.37	3.59	0.09	0.63	0.91	0.37	2.42	1.28
5  -6	46	2.75	2.51	0.05	0.63	0.95	0.37	2.26	1.60
6 -7	29	1.74	1.51	0.03	1.00	0.97	0.00	2.00	2.50
> 7	91	5.45	0.96	0.03	0.00	0.97	1.00	1.00	0.73

%n(x): percentage of dogs per age class; d(x): cumulative mortality; e(x): age-class specific life expectance; l(x): cumulative survival; m(x): number of puppies born per female and age-class; n(x): number of dogs per age class; p(x): age-class specific survival; q(x): age-class specific mortality.

Figure 2

Survivorship and mortality curves for the studied canine population.



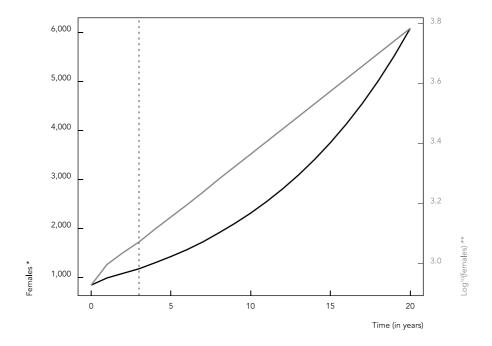
was very small (2.75 years), with the highest life expectancy observed in age-class 1 to 2 years. The greatest average number of viable offspring per female was found in ages between 6 and 7 years, estimated at 2.5 puppies.

Using the age-classified Leslie matrix model, we estimated that the growth rate and the intrinsic rate of increase in the studied dog population was 1.10 and 0.10, respectively. Generation time per female was estimated at 3.4 years and re-

placement rate per female (i.e., net reproductive rate) was estimated at 1.45 offspring per female per generation. The highest reproductive value was observed between 4 to 5 years of age. The projection of population growth over time resulted in stable ratios between age classes after three iterations, and the simulated structured popula-

Figure 3

Simulation of canine population growth in Panorama, São Paulo, Brazil.



<sup>\*</sup> Population growth projection in absolute number of females;

tion grew exponentially after the third simulated year (Figure 3). Additionally, from 1 to 7 years of age, reproductive values ranged from 2.29 to 3.32 future puppies per female, with age class 4 to 5 years being the most fertile.

## Discussion

We sought for information on the structure and growth potential of a dog population under high culling rate due to leishmaniasis challenge, a very frequent situation in many Brazilian urban areas.

The observed dog to human ratio in Panorama (1:7) was similar to the one observed in Araçatuba, São Paulo State (1:8), an area that is also VL endemic thus where euthanasia is compulsory <sup>16</sup>. Additionally, the ratio was smaller than previously estimated (1:4) for 41 municipalities in São Paulo State <sup>17</sup>, or for São Paulo city <sup>18</sup>, which may reflect the lower population density observed in Panorama (0.31 dogs per household and 1 dog per dog-owning household) and the predominance of houses instead of vertical apartment buildings.

The population in the study was young with 50.7% of the dogs aged less than one year, and the expansive pattern observed in the Panorama age pyramid (Figure 1) is consistent with high birth and death rates. Additionally, replacement and migration may have contributed to the wider base for the first age class. Owners have a preference for adopting puppies 10 and Andrade et al. 16 observed that, after an increase in the dogculling rate in response to the advent of the CVL endemics in Araçatuba, the percentage of puppies aged less than one year increased from 20% to 32.5%, possibly due to dog replacement. In contrast, the replacement rate reported by owners in this study was 7.8% (93/1,194) and the observed migration rate was 3.2%. Although health agents had been trained to check the informed dog's age, this may result from biased information provided by the owners, and a longitudinal study would be necessary to better evaluate the influence of immigration and emigration to the age structure of this population 19.

Many causes may have contributed to the high mortality (67%) in the first age-class, including the occurrence of parasitic and infectious

<sup>\*\*</sup> Population growth projection in a logarithmic scale.

diseases, malnutrition and bad habitat conditions 20,21, a very common scenario found in dog populations in Brazil. Responsible pet ownership must be stimulated to enhance the health status of dog populations, particularly at young ages. Feeding their dogs properly, providing them with a clean place to live, vaccinating them regularly and preventing them from roaming freely, are practices that owners must be aware of when they adopt their pets.

For the following age classes, compulsory culling may have influenced the mortality of the dog population and contributed to the high cumulative mortality in next age classes 2 to 3 and 3 to 4 years (Table 1). Although information on the age of culled dogs was not available for the studied population, dogs from Araçatuba were more frequently culled at the age of 34 months 10. This is consistent with the expected life span of 2.75 years (33 months) estimated for dogs from Panorama. The age structure of the population surveyed in our study is also remarkably similar to that reported by Gsell et al. 19 for Iringa, Tanzania, with a high mortality rate and young dog age structure where rabies is endemic. This is an important aspect that should be considered when planning additional control measures such as birth control by mass sterilization, since the success of such measure is directly influenced by the age structure of the target population.

Of note, in spite of the high culling rate applied to this population, the growth projection analysis (Figure 3) showed that under a constant environment and fixed survival and fecundity, the population has potential to grow in a logarithmic scale over the years. A similarly structured dog population from Iringa, Tanzania, where rabies is a challenge, was reported to have the same growth behavior 20. In regard to CVL, the absence of birth control, in the long term, could contribute to the maintenance of VL. Implementation of mass sterilization programs, in this scenario, could be of interest.

Parameters estimated in the present study can be useful for mathematical modeling that have been proposed to simulate the effect of surgical sterilization on canine populations 22,23 and will help local authorities to have a cost-effect sterilization program. For the studied dog population, for instance, it seems that better results would be achieved if dogs aging from 1 to 3 years were targeted, due to its specific survival, fecundity and life expectancy in spite of compulsory euthanasia. Although age miss assignments in adult dogs were expected in our study, these were unlikely to significantly bias our results, since the largest contingency of animals here were very young and could not be mistaken with adult animals.

#### **Conclusions**

An owned-dog population under compulsory culling in a VL endemic area tends to be young, with a low life span. However, in spite of the ongoing process of elimination and renewal, the canine population still has the potential to grow over the years. The estimated parameters can be further applied in models to maximize the impact and minimize financial inputs of alternative measures for VL control.

# Contributors

D. V. Bortoletto collected and analyzed the data and wrote the paper. Y. T. Utsunomiya analyzed the data and wrote the paper. S. H. V. Perri analyzed the data. F. Ferreira conceived and designed the study and analyzed the data. C. M. Nunes conceived and designed the study, wrote the paper, and coordinated the research. All authors read, approved and contributed to the editing of the final manuscript.

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#### Conflict of interests

The authors declare that they have no competing interests.

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#### Resumo

É importante conhecer a estrutura etária da população canina para melhor planejar e avaliar os programas de controle de zoonoses. Os autores analisaram os dados de um censo de cães com donos para caracterizar, pela primeira vez, a estrutura etária de uma população canina sujeita à eliminação compulsória numa área endêmica de leishmaniose visceral (Panorama, Estado de São Paulo, Brasil) que registrou uma taxa de eliminação canina de 28% no ano do estudo. Os dados para 1.329 domicílios e 1.671 cães com donos mostraram uma razão de cães para humanos de 1:7. A média de idade dos cães foi estimada em 1,73 anos; a pirâmide etária indicou altas taxas de natalidade e de mortalidade no primeiro ano de vida, com uma estimativa de mortalidade cumulativa de 78% aos três anos de idade, e uma expectativa de vida de 2,75 anos. Apesar da alta mortalidade, uma simulação de projeção de crescimento sugeriu que a população tem o potencial de crescer numa escala logarítmica ao longo dos anos. Os parâmetros estimados podem ser aplicados também a modelos para maximizar o impacto e minimizar os insumos financeiros de medidas de controle da leishmaniose visceral.

Cães; Distribuição por Idade; Leishmaniose Visceral; Zoonoses

#### Resumen

El conocimiento de la estructura etaria de una población de perros es esencial para la planificación de programas de control de zoonosis. Se analizaron datos de un censo de población canina domiciliada, con el objetivo de caracterizar, por primera vez, la estructura de una población de perros domiciliados en un área donde la eutanasia de perros positivos en leishmaniasis visceral es obligatoria (Panorama, São Paulo, Brasil), y que registró un 28% de casos de eutanasia en el año en que el censo fue realizado. Los datos de 1.329 domicilios y 1.671 perros resultaron en una razón perro:hombre de 1:7. La edad media de los perros fue estimada en 1,73 años; la pirámide de edad indica altas tasas de nacimiento y mortalidad hasta 1 año de vida, con tasa de mortalidad acumulada de un 78% en el tercer año de vida, y expectativa de vida de 2,75 años. A pesar de la alta tasa de mortalidad, la simulación de crecimiento poblacional sugiere que esta población tiene potencial de crecimiento en escala logarítmica a lo largo de los años. Los parámetros estimados pueden ser utilizados en modelos para maximizar el impacto y minimizar los costes de las medidas de control de la enfermedad.

Perros; Distribución por Edad; Leishmaniasis Visceral; Zoonosis

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