

Main Measures Of Hemoparasite Morbidity In Dogs Using Idexx Snap 4dx At The Veterinary Teaching Hospital Of The San Francisco University Of Quito.

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Summary

Background: Hemoparasitism is a vector-related disease that produces many clinical signs. There are diagnostic ELISA tests such as the Idexx snap 4Dx test. In order for the disease to develop, it requires certain aspects of the etiological agent, the host and the environment.

METHODOLOGY: This is a retrospective, observational, longitudinal, epidemiological study, which aims to study the main morbidity measures of hemoparasites in dogs. The quantitative variables were analyzed: body temperature, hematocrit level, and age. The qualitative variables were sex, breed, presence of ectoparasites in the dog, and trips by the dog to the Ecuadorian coast. By determining central tendency measures (MTC), frequency tables, contingency tables with the establishment of the Odds Ratio (OR) as the association unit and statistical significance by determining the Chi Square and the strength of association of differences in means and statistical significance by means of the Student T statistical test. For this study, a 95% confidence interval and a 5% margin of error were used. At the end, an endemic channel was made.

Results: The prevalence of hemoparasites in the Veterinary Teaching Hospital of the San Francisco University of Quito is 16.89%. The main etiological agent of hemoparasites is *Ehrlichia canis* with 56% of positive cases recorded. 32% of cases are due to *Anaplasma phagocytophilum*. Three cases of *Dirofilaria immitis* are recorded, which is equivalent to 12%. No positive case for *Borrelia burgdorferi* was found. The central tendency measures for the average body temperature is $39.55^{\circ}\text{C} \pm 0.46$. The average hematocrit is $42.29\% \pm 9.58$. The age has an average of $3.70 \text{ years} \pm 3$. Using the different averages of the quantitative variables, it was determined that the hematocrit level has an average of 42.8% with a statistical T of 0.74 and a $p = 0.74$. For the body temperature an average of 39.67°C with a statistical T of 1.36 and a $p = 0.17$. The age does not present a relationship so a statistical analysis cannot be performed. The qualitative variables were analyzed and their results are: the variable sex was observed that females have $\text{OR} = 1.58$, which did not represent statistical significance. The variable race was observed that crossbreeds have an $\text{OR} = 2.60$ and is statistically significant. The variable presence of ectoparasites (ticks) has an $\text{OR} = 7.11$ and is statistically significant. For the variable trips to the Ecuadorian coast there is an $\text{OR} = 8.21$ and is statistically significant. The endemic channel in its security zone has peaks in the months of April, September and October of hemoparasites with 1 positive case per month. **Conclusions:** There is evidence that dogs in the Ecuadorian highlands are at risk of becoming infected with hemoparasites through vector transmission, as the risks of transmission are unknown. Although exposure may vary depending on the agent and the geographic region traveled to, although not yet described by control bodies, the probability of infection with at least one agent is considerable throughout the Ecuadorian coast.

Keywords: Ehrlichiosis, Anaplasmosis, Borreliosis, Dirofilariosis, vectors, hemoparasites, endemic channel, odds ratio.

BACKGROUND

In this retrospective study, hemoparasitism includes the diseases Ehrlichiosis, Anaplasmosis, Borreliosis and Dirofilariosis, which are linked to vectors. It is a disease that has many clinical signs that, when combined, can be related to many diseases and different diagnoses. Currently, there are commercial ELISA tests such as the Idexx snap 4Dx test, which is a rapid diagnostic aid that can be used to detect the presence or absence of any of the etiologic agents with the patient's serum, plasma or whole blood. The reagent detects antigens for *Dirofilaria immitis* (canine heartworm) and antibodies against *Anaplasma phagocytophilum*, *Borrelia burgdorferi* and *Ehrlichia canis*. A correct diagnosis helps to ensure an effective and safe treatment for the pet. Hemoparasitism requires aspects of the etiological agent, the host and the environment, which can only occur through their relationship. To analyze the strength of association between qualitative and quantitative variables, risk factors are evaluated. For this retrospective study, it was performed through the odds ratio, which gives us an idea of how much exposure one group has to the other, what the relationship is between both groups and the probability of presenting hemoparasitism in these can be compared, at the Veterinary Teaching Hospital of the San Francisco University of Quito. Knowing how many cases of hemoparasites exist is important in order to estimate the probability of how many positive cases of hemoparasites will occur in a period of time and to be able to compare and analyze what happens with hemoparasitism in different places in the country and the world. This study carried out an endemic channel that proposes measuring the presence of hemoparasitism from 2011 to 2015 and thus having the necessary tools to manage the relevant measures, which not only affect animal welfare but also human beings.

GOALS

General objective

To determine the main morbidity measures of hemoparasites in dogs (*Canis familiaris*) through the Idexx 4Dx snap from 2011 to 2015 at the Veterinary Teaching Hospital of the San Francisco University of Quito"

Specific objectives

1. To determine the prevalence of hemoparasites in dogs from 2011 to 2015 at the Veterinary Teaching Hospital of the Universidad San Francisco de Quito.
2. To determine the association measures between the presence of hemoparasites with: sex, age, breed, hematocrit level, presence of ectoparasites in the dog, body temperature and the dog's travels to the Ecuadorian coast.
3. Establish the endemic channel for hemoparasitism at the Veterinary Teaching Hospital of the San Francisco University of Quito.

METHODOLOGY

It is a retrospective observational longitudinal epidemiological study, which aims to study the main measures of morbidity of hemoparasites in dogs, with the prevalence indicator that allows to estimate and observe the probability of finding positive cases of hemoparasites in dogs in the Veterinary Teaching Hospital of the San Francisco University of Quito. Descriptive statistics were used for the study, with a total of 148 samples of the Idexx snap 4 Dx from 2011 to 2015 in an Excel database, the qualitative and quantitative variables, with a significance level of 95% and a margin of error of 5% were analyzed in the statistical program Epi Info 7, with which the frequencies were determined and analyzed using graphs and comparative tables. In addition, the association measures evaluated for the univariate analysis were performed for quantitative variables (body temperature, hematocrit level and age), by determining measures of central tendency (MTC): mean, median and standard deviation. For qualitative variables (sex, breed, presence of ectoparasites in the dog and trips of the dog to the Ecuadorian coast) frequency tables were made. The bivariate analysis comparing the probability of the occurrence of hemoparasitism in two groups of qualitative-quantitative variables was performed using contingency tables with the establishment of the Odds Ratio (OR) as the association unit and statistical significance by determining the Chi Square. The quantitative-qualitative bivariate analysis was performed using the strength of association of differences in means and statistical significance by means of the Student T test.

To create an endemic index that uses the frequency of cases of hemoparasitism per month that have occurred between 2011 and 2015 at the Veterinary Teaching Hospital of the Universidad San Francisco de Quito, and thus create an endemic channel that serves to determine the regular frequency of hemoparasites in dogs at the Veterinary Teaching Hospital of the Universidad San Francisco de Quito and predict their behavior over time and, at some point, be able to make decisions in the event of a risk of an outbreak.

RESULTS AND DISCUSSION.

The result of the prevalence of hemoparasites in the Veterinary Teaching Hospital of the San Francisco University of Quito is 16.89% (See table 1). This result compared with another city in the Ecuadorian highlands such as Cuenca, according to Dominguez. He reports in his thesis that it has a prevalence of 11.43% (Dominguez Alvarez, 2011, pág. 62). In the city of Milagro, Ecuadorian coast, Marquez points out in his thesis research that it has a frequency of 57% (Marquez Cabrera, 2011, pág. 52). In Nicaragua, in district VII 2 of the city of Managua, Angulo & et al explain that there is a prevalence of 17.73%. (Angulo Campos, Rodríguez Vilchez, Hernández, & Fonseca, 2005, pág. 70)

Table 1 Prevalence of hemoparasites.

Result	Frequency	Percentage	Cumulative Percentage
Negative	123	83.11%	83.11%
Positive	25	16.89%	100.00%
Total	148	100.00%	100.00%

Source: Prepared at the Veterinary Teaching Hospital of the San Francisco University of Quito.

The result of positive cases indicates that the main etiological agent of hemoparasites is *Ehrlichia canis*, with 56% of positive cases registered (See table 2). According to Dominguez, in the city of Cuenca *Ehrlichia canis* is the main etiological agent for hemoparasites with 56.25% (Dominguez Alvarez, 2011, pág. 5). Marquez points out that *Ehrlichia canis* has 43.85% as a single etiological agent and 40.35% with mixed infections with Anaplasmosis (Marquez Cabrera, 2011, pág. 74). Angulo & et al explain that Managua has a lower prevalence of *Ehrlichia canis*, with 4.6%. Being a Caribbean area, it has other etiological agents such as *Haemobartonella canis*, *Babesia canis* and *Hepatozoon canis* with a higher prevalence described in this study (Angulo Campos, Rodríguez Vilchez, Hernández, & Fonseca, 2005, pág. 70). Cardoso & et al describe that in Portugal the healthy group has 4.1% and the suspected group 16.4% (Cardoso, Mendão, & Madeira de Carvalho, 2012, pág. 1).

Table 2 Percentage of positive cases.

SPECIES +	Frequency	Percentage	Cumulative Percentage
<i>Ehrlichia canis</i>	14	56.00%	56.00%
<i>Anaplasma phagocytophilum</i>	8	32.00%	88.00%
<i>Dirofilaria immitis</i>	3	12.00%	100.00%
<i>Borrelia burgdorferi</i>	0	0%	100.00%
Total	25	100.00%	100.00%

Source: Prepared at the Veterinary Teaching Hospital of the San Francisco University of Quito.

32% of positive cases are due to *Anaplasma phagocytophilum* (See Table 2). Márquez points out in his study a 15.78% and 40.35% with mixed infections with Ehrlichiosis (Marquez Cabrera, 2011, pág. 71). Of the same in the city of Cuenca, Domínguez explains in his study a 3.13% (Domínguez Alvarez, 2011). While Cardoso & et al points out that the healthy group covers 4.5% and in the suspected group 9.2% (Cardoso, Mendão, & Madeira de Carvalho, 2012, pág. 1). Mejía & Calero in the urban parish of Portoviejo, province of Manabí, a 13.10% (Mejía Chica & Calero Carreño, 2013, pág. 1). There are three cases of *Dirofilaria immitis*, which is equivalent to 12% (see table 2). Marquez says that no positive cases were found (Marquez Cabrera, 2011, pág. 74). Cardoso et al. found in their study that the healthy group has 3.6% and the suspected group 8.9% (Cardoso, Mendão, & Madeira de Carvalho, 2012, pág. 1). Díaz and Santiago in the municipality of Sucre, state of Sucre in Venezuela, report 15.2%. (Díaz & Santiago, 2011, pág. 51) No positive case for *Borrelia burgdorferi* was found at the Veterinary Teaching Hospital of the Universidad San Francisco de Quito (See Table 2). In the city of Milagro, no case was recorded either, as analyzed by Márquez (Marquez Cabrera, 2011, pág. 74). Rubio et al. report 2 cases for the first

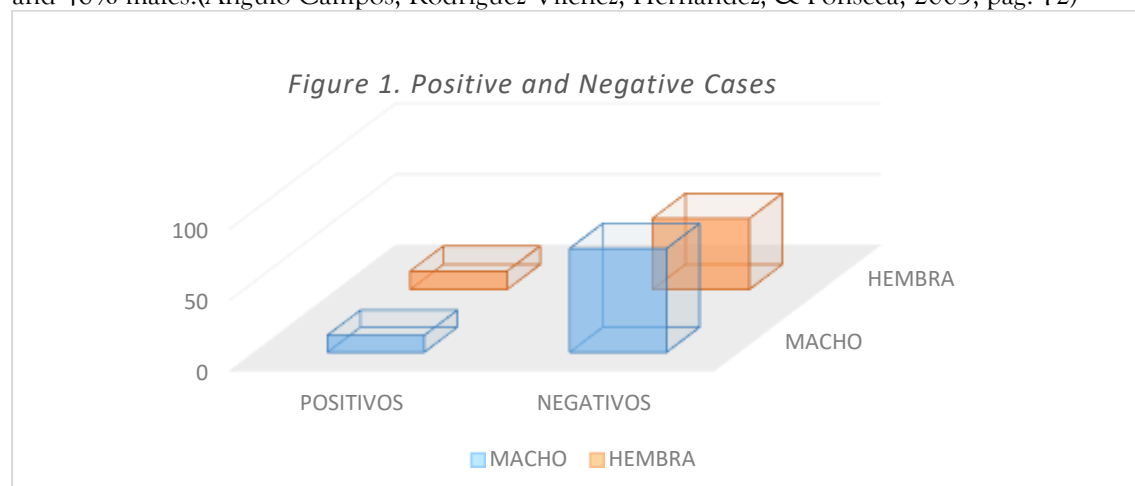
time in the city of Lima, Peru, the diagnosis was made with a commercial snap 4Dx ELISA test (Rubio, Salas, & Gómez, 2011). The quantitative variables whose results of the central tendency measures are: the average for body temperature is 39.55 ° C. Fever was the first sign of consultation at the Hospital with peaks of 41 ° C and has a standard deviation of 0.46 (See table 3). Angulo & et al in Managua investigated that the average body temperature was 39.45 ° C and has a standard deviation of 1.49 (Angulo Campos, Rodríguez Vilchez, Hernández, & Fonseca, 2005, pág. 75). There are no studies at the national level that have taken into account body temperature, therefore it is a new contribution to these studies of hemoparasites in dogs.

Table 3 Univariate analysis, quantitative variables of hemoparasitism.

MTC	Body Temperature (°C)	Hematocrit Level (%)	Age (Years)
Average	39.55	42.29	3.70
Median	39.60	44	3
Standard deviation	0.46	9.58	3.00
Minimum	38.2	9	0.1
Maximum	41	62	12

Source: Prepared at the Veterinary Teaching Hospital of the San Francisco University of Quito.

The average hematocrit of the study was 42.29% and has a standard deviation of 9.58, which is within the normal range of the research. These are contrasted with the results of the literature review, which in terms of symptoms say that anemia should exist (See Table 3). Angulo & et al in Managua reported an average of 29.88% and has a standard deviation of 9.93 (Angulo Campos, Rodríguez Vilchez, Hernández, & Fonseca, 2005, pág. 76). There are no studies at the national level that have taken into account the hematocrit level in the dogs sampled, therefore it is a new contribution to these studies of hemoparasites in dogs. The age has a standard deviation of 9.58 and an average of 3.70 years, which tells us that the dogs that come to the consultation are young and have a standard deviation of 3 (See Table 3). Of the total cases of hemoparasites in dogs from 2011 to 2015 at the Veterinary Teaching Hospital of the San Francisco University of Quito, the negative cases were 123. Of 25 positive cases of hemoparasites, we have a sex ratio with 52% females and 48% males (See figure 1). According to Márquez in the city of Milagro, the ratio is: 53% are females and 47% are males in positive cases (Marquez Cabrera, 2011, pág. 74). Domínguez in the city of Cuenca explains that males have 65% and females 35% of positive cases (Domínguez Alvarez, 2011, pág. 5). Likewise, Angulo & et al in Managua report that 54% were females and 46% males. (Angulo Campos, Rodríguez Vilchez, Hernández, & Fonseca, 2005, pág. 72)



Source: Veterinary Teaching Hospital of the San Francisco University of Quito.

They were classified into two groups: those with phenotypic characteristics of certain breeds in pure form and those with mixed breeds, which are a mixture of these breeds, without racial characteristics. This gave a total of 84.46% pure breed patients and 15.54% mixed breed patients (see table 4).

Table 4 Univariate analysis .

	Classification	Frequency	Percentage	Cumulative Percentage
Race	PURE	125	84.46%	84.46%
	MESTIZO	23	15.54%	100.00%
	TOTAL	148	100.00%	100.00%
Ectoparasites	NO	49	33.11%	33.11%
	YEAH	99	66.89%	100.00%
	TOTAL	148	100.00%	100.00%
Trips to the Ecuadorian coast	NO	68	45.95%	45.95%
	YEAH	80	54.05%	100.00%
	TOTAL	148	100.00%	100.00%

Source: Veterinary Teaching Hospital of the San Francisco University of Quito.

Of 148 dogs that attended the medical consultation, 66.89% of them had ticks on their tegument (See table 4). Angulo et al in Managua reported that the presence of ticks is 58.64% of the total sample. (Angulo Campos, Rodríguez Vilchez, Hernández, & Fonseca, 2005, pág. 98)The percentage of trips to the Ecuadorian coast of dogs that accompany their owners is 54.05%. (See table 4) The hematocrit level in patients with the presence of hemoparasites was found to be within the normal average, that is, 42.8% (see table 4). No statistically significant difference was observed. Comparing this with the study by Angulo et al in Managua, the presence of anemia was found with 27.89% of the hematocrit level (Angulo Campos, Rodríguez Vilchez, Hernández, & Fonseca, 2005, pág. 76). This discrepancy can be explained as follows: Peña explains that hypobaric hypoxia is a stress factor due to living at altitude and stimulates morphological and physiological adaptations (hematological and circulatory) to cope with the lower concentration of atmospheric oxygen (Peña, 2011, págs. 4-6). In addition, Angulo et al. points out that in Managua, hematocrit levels can be influenced by factors such as: management, nutrition, high loads of ectoparasites and endoparasites (Angulo Campos, Rodríguez Vilchez, Hernández, & Fonseca, 2005, pág. 75). For body temperature, it was found that a dog with hemoparasites is 1.36 times more likely to have a fever than a dog without hemoparasites. No statistically significant difference was found (see table 5). There is no relationship between the presence of hemoparasites and age, so a statistical analysis cannot be performed. The qualitative variables gave the following results: the sex variable showed that females were 1.58 times more likely to contract hemoparasites than males, this result was not statistically significant. (See table 6) For the variable race it was observed that a mixed breed has 2.6 times more probability of contracting hemoparasites than pure breeds. This result was statistically significant. (See table 6) According to Escalante in his study of the evaluation of the purchase of dogs in the city of Guayaquil, he says that purebred dogs are bought at exorbitant prices therefore, the market for these pets is growing, breeders sell dogs for their appearance and the population supports this form of sale (Escalante Cirino, 2013, págs. 2-6). As a result of this observation it is deduced that it is a matter of taste and economic power where the mixed breed is frowned upon and could be related to the fact that it requires less care and attention.

Table 5 Bivariate analysis, quantitative variables.

		Observed	Average	Median	Standard deviation	T statistic	P value
Hematocrit level	Positive for hemoparasites	25	42.8	45	8.27	0.33	0,74 ^{ns}
	Negative for hemoparasites	123	42.1	44	9.85		
	Positive for hemoparasites	25	39.67	39.70	0.57	1.36	0,17 ^{ns}

Body temperature	Negative for hemoparasites	123	39.53	39.60	0.44	0.73	0,46 ^{ns}
	Positive for hemoparasites	25	4.10	3	23		
Age	Negative for hemoparasites	123	3.62	3	3.11		

ns. Not significant.

Source: Prepared at the Veterinary Teaching Hospital of the San Francisco University of Quito.

For the variable presence of ectoparasites (ticks) it was observed that there is a 7.11 times greater probability of contracting hemoparasites in dogs that have ticks compared to those that do not have ticks. (See table 6) This result is statistically significant and could be corroborated with that stated by Cardoso & et al. who found an OR = 2.7 with a p value of 0.185 (Cardoso, Mendão, & Madeira de Carvalho, 2012, pág. 7). There are no studies at the national level that have taken into account the presence of ectoparasites (ticks), which is why it means a new contribution to these studies of hemoparasites in dogs. This is explained because ticks are vectors that carry diseases such as: *Anaplasma phagocytophilum*, *Borrelia burgdorferi* and *Erhlichia canis*.

Table 1 Bivariate analysis, qualitative variables.

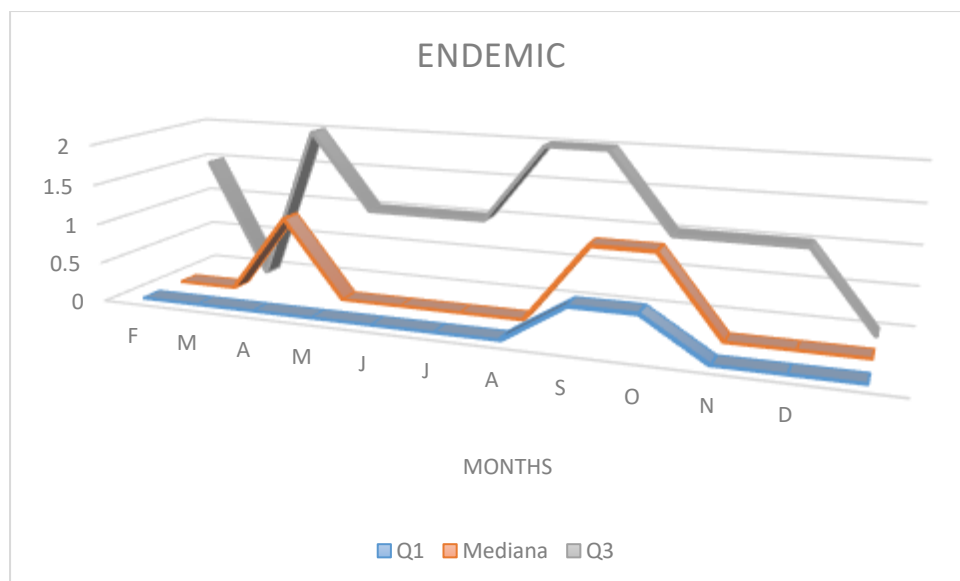
		Hemoparasites		OR value	95% confidence intervals	Chi square
		Positives	Negatives			
Sex	Female	13	50	1.58	0.66-3.74	0,15 ^{ns}
	%	20,63	79.37			
	Male	12	73	2,6007	0.93-7.20	0.040 ^s
	%	14,12	85.88			
Race	Mestizo	7	16	7,11	1.60-31.55	0.0010 ^s
	%	30,43	69.57			
	Pure	18	107	8.21	2.33-28.89	0.000062 ^s
	%	14,40	85.60			
	YEAH	23	76			
	%	23,23	76,77			
Ectoparasites	NO	2	47			
	%	4.08	95.92			
	YEAH	22	58			
	%	27,50	72,50			
Trips to the Ecuadorian coast	NO	3	65			
	%	4,41	95.59			

ns. Not significant.

s. Significant.

Source: Prepared at the Veterinary Teaching Hospital of the San Francisco University of Quito.

For the variable trips to the Ecuadorian coast, it was observed that there is an 8.21 times greater probability of contracting hemoparasites in dogs that travel compared to those that stay in the Ecuadorian mountains. This result is statistically significant (See Table 6). Cardoso et al recommend having updated information on the epidemiology of hemoparasitosis to assign the regional risk (Cardoso, Mendão, & Madeira de Carvalho, 2012, pág. 3). With the results presented, there is a correlation between hemoparasitosis and the geographic area.



Source: Prepared at the Veterinary Teaching Hospital of the San Francisco University of Quito.

The endemic channel of the Veterinary Teaching Hospital of the Universidad San Francisco de Quito has a median endemic index. In its security zone, it has peaks in the months of April, September and October of hemoparasites with 1 positive case per month. It also has a relationship with the school schedule of the mountains and the Amazon of the Ministry of Education of Ecuador; which places its vacations of the first quarter at the end of February and the vacations for the end of the school year in the months of July to September (Educación, 2015, pág. 1). Time in which the whole family, including the dog, enjoys the vacations, that is, traveling and visiting cities, beaches and other wonderful places that the Ecuadorian coast offers.

CONCLUSIONS

The present study provides evidence that dogs in the Ecuadorian highlands are at risk of becoming infected with hemoparasites through vector transmission, as the risks of transmission are unknown. Although exposure may vary depending on the agent and the geographic region traveled to, and have not yet been described by control bodies, the probability of infection with at least one agent is considerable throughout the Ecuadorian coast.

Prophylactic measures are based on controlling vector density, including the use of ectoparasiticides, as well as knowing the characteristics of the etiological agent to protect dogs and simultaneously limit the risk of zoonotic transmission. Finally, it is expected that public health authorities will have a greater concern about the image of hemoparasitism in Ecuador in order to establish future control programs.

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