

# Molecular detection of *Hepatozoon* species in domestic cats and dogs living in Germany (2007-2021)

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## INTRODUCTION

*Hepatozoon* (*H.*) species are protozoal infectious pathogens transmitted by blood-feeding arthropods.<sup>1,2</sup> Three *Hepatozoon* species (*H. felis*, *H. canis* and *H. silvestris*) have been reported to infect cats, whereas only *H. canis* is known to infect dogs in Europe.

## MATERIAL AND METHODS

The study included cats and dogs tested for *Hepatozoon* species by PCR between 2007 and 2021 by the LABOKLIN laboratory. The submitting veterinarians were asked if the tested animals had stayed abroad. From 2018 to 2020, a partial 18S rRNA *Hepatozoon* gene fragment was sequenced from cats that tested positive. Descriptive statistical analysis was carried out using SPSS for Windows (version 28.0; IBM).  $P < 0.05$  was regarded as statistically significant.

## OBJECTIVES

The objective of this study was to determine the prevalence of *Hepatozoon* species in samples from cats and dogs living in Germany and to identify potential risk factors for infection.

**Table 1:** Age distribution in dogs and cats tested by *Hepatozoon* species PCR at LABOKLIN from 2007 to 2020 (n tested positive/N total (%))

Age	Dogs	Cats
Puppy/kitten (≤ 6 months)	280/4,814 (5.8)	7/50 (14.0)
Junior (> 6 months to 3 years)	1,985/51,299 (3.9)	35/554 (6.3)
Adult (> 3 – 7 years)	720/16,775 (4.3)	14/203 (6.9)
Mature (> 7 – 10 years)	378/10,453 (3.6)	10/110 (9.1)
Senior (> 11 – 14 years)	90/3,121 (2.9)	2/66 (3.0)
Geriatric (≥ 15 years)	11/278 (4.0)	0/24 (0)
<b>Total</b>	<b>3,464/86,740 (4.0)</b>	<b>68/1,007 (6.8)</b>

**Table 2:** Sex of dogs and cats tested by *Hepatozoon* species PCR at LABOKLIN from 2007 to 2020 (n tested positive/N total (%))

Sex	Dogs	Cats
Male	684/20,158 (3.4)	7/133 (5.3)
Male castrated	876/22,860 (3.8)	31/452 (6.9)
Female	1,005/22,844 (4.4)	10/123 (8.1)
Female spayed	1,280/28,525 (4.5)	20/387 (5.2)
<b>Total</b>	<b>3,845/94,387 (4.1)</b>	<b>68/1,095 (6.2)</b>

## RESULTS

In total, 76/1,186 cats (6.4%) and 4,335/102,037 dogs (4.2%) tested positive for *Hepatozoon* spp. The percentage of dogs tested positive was over 5% in the last three years (Figure 1), whereas in cats varying percentages are seen (Figure 2). Stays abroad were reported in all animals that tested positive and for which an anamnesis could be obtained. The 18S rRNA gene of all 16 sequenced feline samples showed 98.7-99.7% identity with a *H. felis* isolate from Spain.

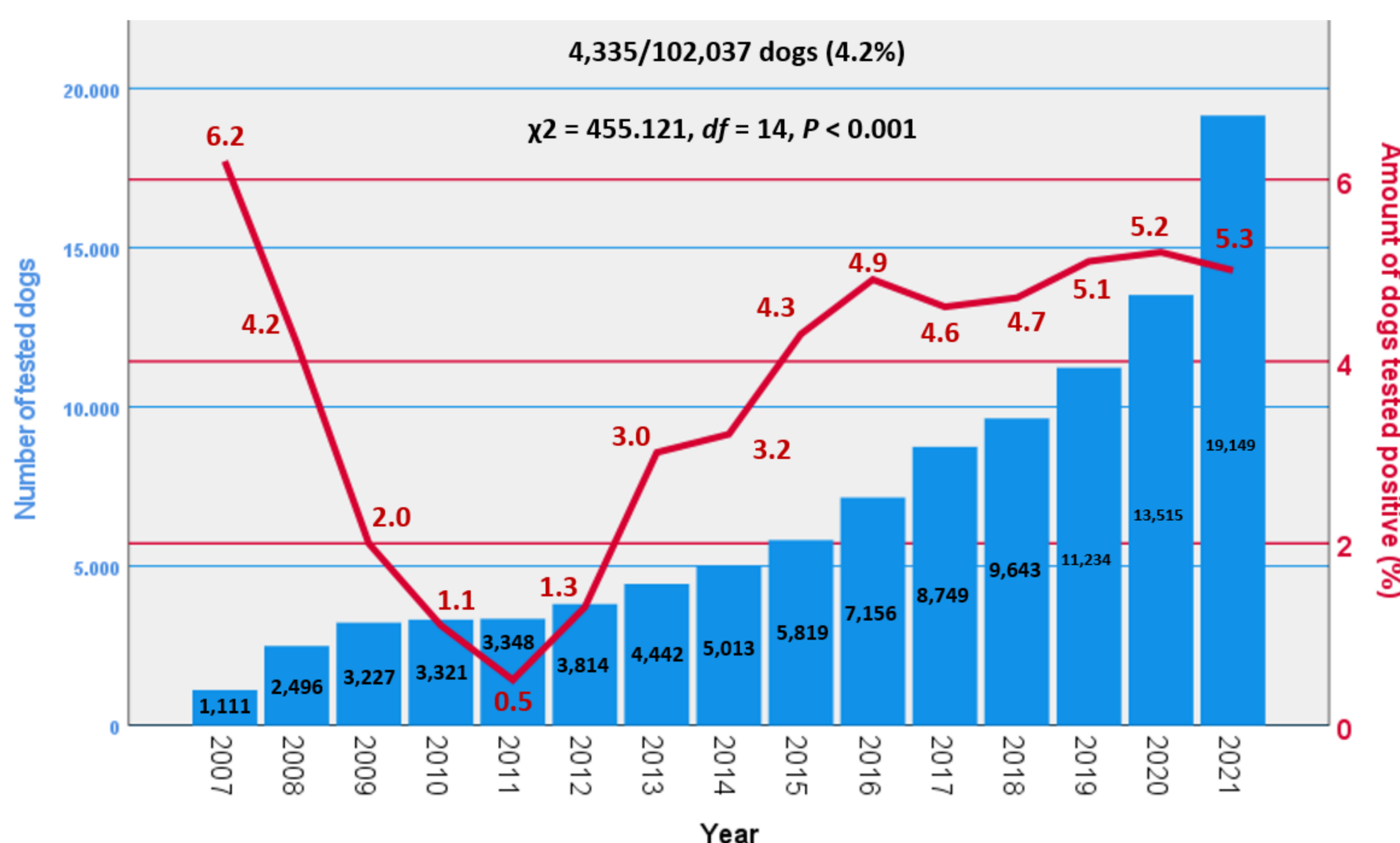
Age was known in 86,740 out of 102,037 dogs (85%) and 1,007 out of 1,186 cats (84.9%) (Table 1). There was a highly statistically significant impact of different age groups in dogs ( $\chi^2 = 61.589$ ,  $df = 5$ ,  $P < 0.001$ ), but not in cats ( $\chi^2 = 8.489$ ,  $df = 5$ ,  $P = 0.131$ ). Sex was known in 94,387/102,037 dogs (92.5%) and 1,095/1,186 cats (92.3%) (Table 2). There was a statistically significant impact on test results in dogs (2,285/51,369 female [4.4%]; 1,559/43,018 male [3.6%]; Fisher's exact test  $P < 0.001$ ), but not in cats (30/510 female [5.9%]; 38/585 male [6.5%]; Fisher's exact test  $P = 0.708$ ). Stays abroad were reported in 4,002/4,128 dogs (96.9%) and all cats with a known anamnesis, with import being the most prominent finding (Table 3). There was a statistically significant impact of stays abroad on test results in dogs ( $\chi^2 = 20.864$ ,  $df = 3$ ,  $P < 0.001$ ).

**Table 3:** Percentage of dogs and cats with known anamneses of stays abroad tested by *Hepatozoon* species PCR at LABOKLIN from 2007 to 2020 (n tested positive/N total (%))

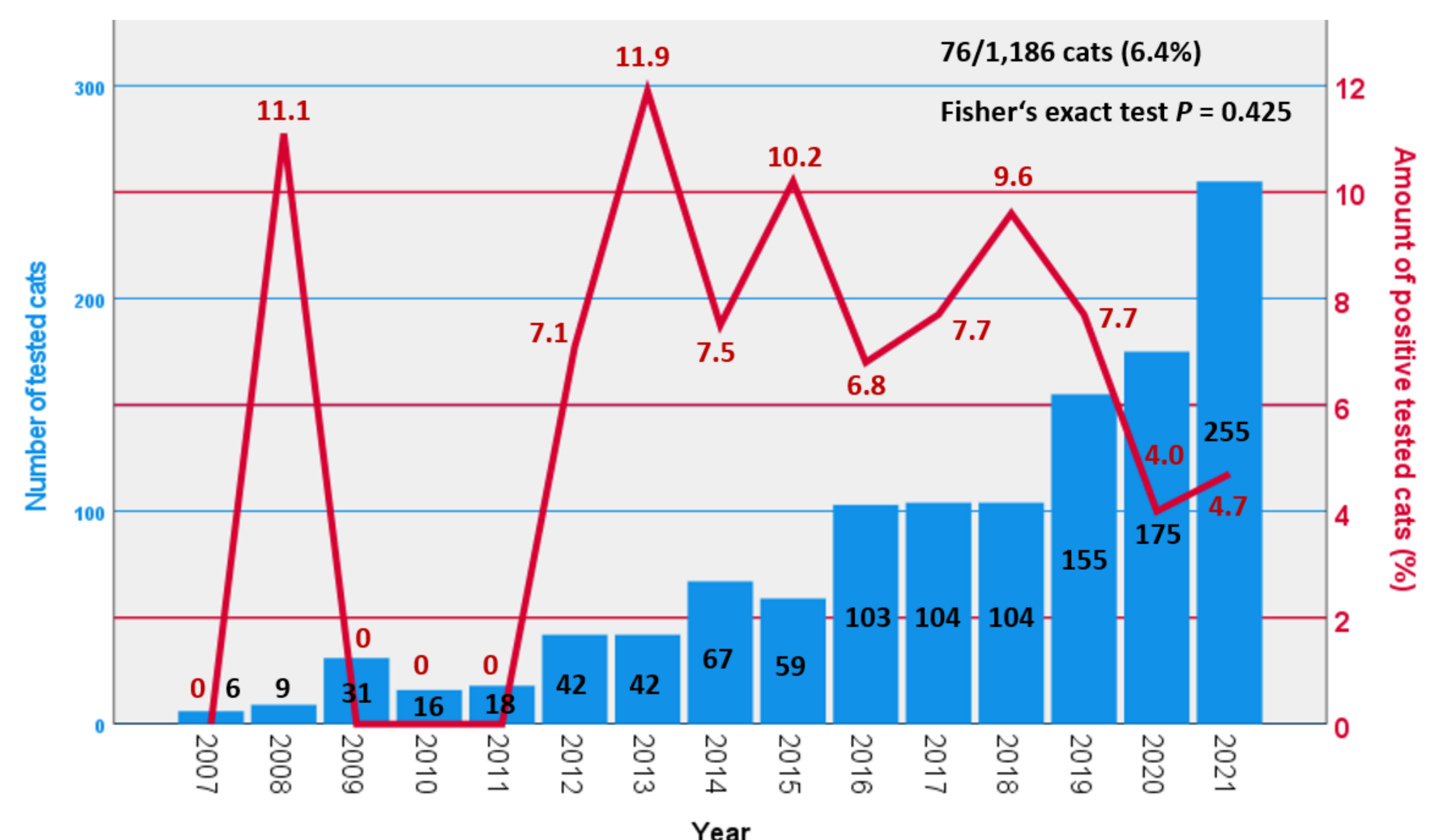
Stays abroad	Dogs	Cats
Import	285/3,782 (7.5)	47/64 (73.4)
Travel	1/136 (0.7)	0/0 (0)
Import and Travel	3/84 (3.6)	0/0 (0)
No import/travel	0/126 (0)	0/0 (0)
<b>Total</b>	<b>289/4,128 (7.0)</b>	<b>47/64 (73.4)</b>

## DISCUSSION and CONCLUSIONS

From 2007-2021, 6.4% of cats and 4.2% of dogs tested positive for *Hepatozoon* spp. In all sequenced feline samples *H. felis* was detected, which agrees with previous reports that this is the main *Hepatozoon* species of domestic cats in Europe.<sup>3</sup> Sex, age and stays abroad were found to be potential risk factors in dogs, but not in cats. However, it must be noted that only a low number of cats was included in this study. Although autochthonous canine and feline *Hepatozoon* infections have been reported in Central Europe<sup>4,5</sup> and infections with *H. canis* are widespread in German fox populations<sup>6</sup>, all positive animals with a known anamnesis included in this study were linked to stays abroad.



**Figure 1:** Results of the *Hepatozoon* species PCR in dogs at LABOKLIN from 2007 to 2021



**Figure 2:** Results of the *Hepatozoon* species PCR in cats at LABOKLIN from 2007 to 2021

## REFERENCES

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