

## INTRODUCTION

Tick-borne encephalitis virus (TBEV) is a member of the genus *Flavivirus* within the family *Flaviviridae* and one of the most important tick-borne diseases in human medicine in Europe. The virus is spread over a wide area of the Eurasian continent. In Germany, it is mainly transmitted by *Ixodes (I.) ricinus* with southern federal states considered as high-risk areas in human medicine.<sup>1,2</sup>

## AIMS OF THE STUDY

Aim of the study was to assess the percentage of positive test results for TBEV in dogs in Germany and to identify possible risk factors for infection/pathogen contact.

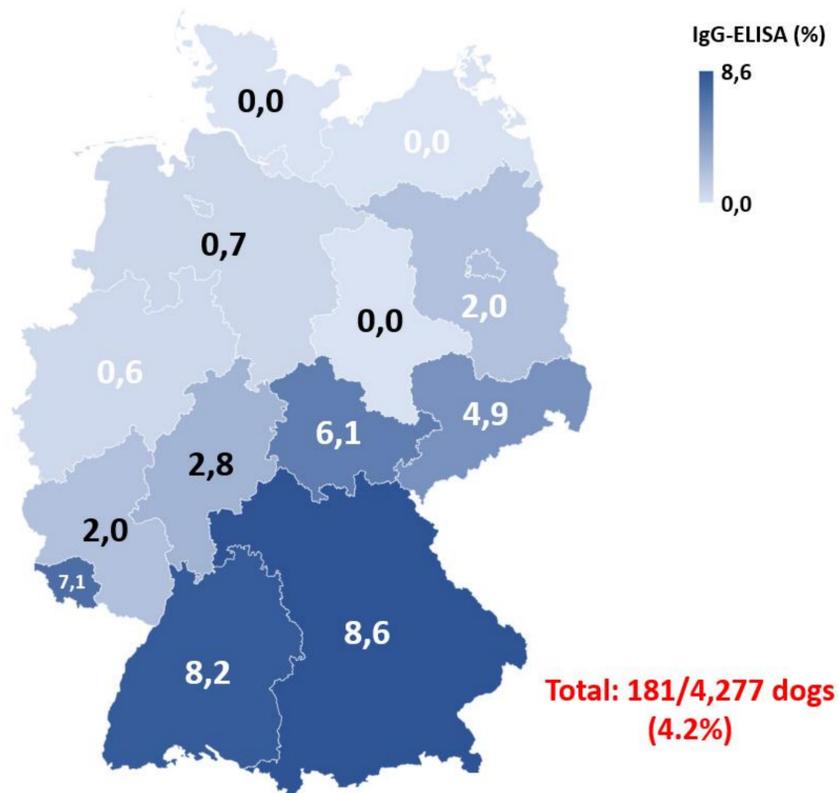


Figure 1: Regional distribution of dogs tested positive for TBEV by IgG-antibody-ELISA testing in Germany from 2010 to 2021 (%)

## MATERIAL AND METHODS

This study evaluated samples taken from dogs between 01/2010 and 12/2021 and analyzed by LABOKLIN (Bad Kissingen, Germany). This included both direct (PCR) and indirect (IgG-all species-ELISA, as well as IgM-ELISA after June 2019) detection methods for TBEV. Only the results of either the first or the first positive test result of each dog were considered.

Table 1: Dogs tested for TBEV by IgG-ELISA from 2010 to 2021

	N tested negative	N tested positive	Total n positive/N total (%)
2010-2014	794	41	41/835 (4.9)
2015-2017	1,978	51	51/2,029 (2.5)
2018-2021	1,324	89	89/1,413 (6.3)
<b>Total</b>	<b>4,096</b>	<b>181</b>	<b>181/4,277 (4.2)</b>

$\chi^2 = 30.622$ ,  $df = 2$ ,  $P < 0.001$

Table 2: Dogs tested for TBEV by IgG-ELISA from 2010 to 2021 sorted by age

Age-group	N tested negative	N tested positive	Total n positive/N total (%)
0 – 2 years (junior)	1,216	28	28/1,244 (2.3)
>2 – 7 years (adult)	1,507	60	60/1,567 (3.8)
>7 – 10 years (mature)	701	44	44/745 (5.9)
>10 – 13 years (senior)	286	25	25/311 (8.0)
>13 years (geriatric)	35	4	4/39 (10.3)
<b>Total</b>	<b>3,745</b>	<b>161</b>	<b>161/3,906 (4.1)</b>

$\chi^2 = 33.147$ ,  $df = 4$ ,  $P < 0.001$

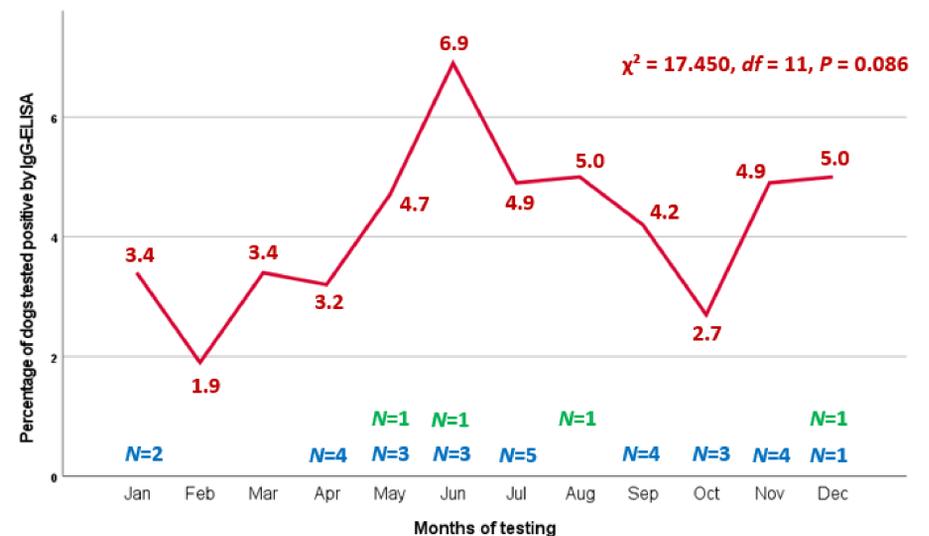


Figure 2: Monthly distribution of dogs tested positive for TBEV by IgG-ELISA (%), IgM-ELISA (N tested dogs) and PCR (N tested dogs) from 2010 to 2021; “Spring” = Mar to May, “summer” = Jun to Aug, “autumn” = Sep to Nov, “winter” = Dec to Feb

## RESULTS

208 out of 4,674 dogs tested positive (5%, PCR 4/401 [1%, liquor n=4]; IgM-ELISA 29/220 [13%, serum n=29]; IgG-ELISA 181/4,277 [4%, serum/plasma n=153, cerebrospinal fluid n=28]). The age was known in 4,259/4,674 dogs (91.1%, median 5.0 years). The pathogen was detected by PCR on cerebrospinal fluid in dogs from Bavaria, Saxony, Lower Saxony, and North-Rhine Westphalia (n=1 each, fig. 2). The rate of dogs with positive serology was highest in southern federal states with statistically significant geographical variation ( $P < 0.001$ , fig. 1). The highest rate of positive IgG-ELISA was seen between 2018 and 2021 (6%,  $P < 0.001$ , tab. 1). Elder dogs had a higher likelihood of being tested positive by IgG-ELISA (OR = 1.976, tab. 2,3). The likelihood of being tested IgG-positive raised 15% each year. In general, the majority of dogs was tested positive in summer for IgG-testing (OR = 1.493, tab. 3, fig. 2).

Table 3: Binominal logistic regression analysis in dogs tested for TBEV by IgG- and IgM-ELISA, all with known sex, age, years as well as months of testing from 2010 to 2021

	B	SE	Wald	P	Odds Ratio	95%-CI for Odds Ratio	
						Lower bound	Upper Bound
<b>IgG- ELISA (N = 3,681 dogs)</b>							
Sex (male)	-0.017	0.170	0.010	0.0921	0.983	0.705	1.372
Age (> 5 years)	0.681	0.170	15.980	< 0.001	1.976	1.415	2.760
Years	0.136	0.033	16.951	< 0.001	1.146	1.074	1.223
Season (summer)	0.401	0.177	5.108	0.024	1.493	1.074	1.223
Constant	-278.182	66.705	17.392	< 0.001	-	-	-
<b>IgM-ELISA (N = 190 dogs)</b>							
Sex (male)	0.192	0.434	0.197	0.658	1.212	0.518	2.835
Age (> 5 years)	-1.056	0.471	5.025	0.025	0.348	0.138	0.876
Years	-0.598	0.321	3.465	0.063	0.550	0.293	1.032
Season (summer)	-0.402	0.464	0.748	0.387	0.669	0.269	1.663
Constant	1206.706	649.001	3.456	0.063	-	-	-

B: unstandardized regression weight; SE: standard deviation to the mean

Variables: male, age > 5 years, year, summer

Degrees of freedom were 1 for all Wald statistics

## LIMITATIONS

There was no data available on ectoparasite prophylaxis, living conditions, possible stays abroad as well as the reasons for testing. This might be of limited importance due to the large number of dogs included in the study.

## DISCUSSION AND CONCLUSIONS

The high percentages of dogs with positive serology between 2018 and 2021 suggest increasing importance of TBEV in dogs in Germany. The geographical distribution of positive serology is consistent with human medicine classifications of high-risk areas.<sup>3</sup> Age, years of testing and months of testing had a statistically significant impact on results of IgG-ELISA testing. Peaks of positive test results were seen in summer.

## REFERENCES

<sup>1</sup>Suss J. Tick-borne encephalitis: Epidemiology, risk areas, and virus strains in Europe and Asia-An overview. *Ticks Tick-Borne Dis.* 2011, 2 1:2-15; <sup>2</sup>Mansfield KL, Johnson N, Phipps LP, et al. Tick-borne encephalitis virus - a review of an emerging zoonosis. *J Gen Virol.* 2009, 90 Pt 8:1781-94; <sup>3</sup>Robert Koch Institut. Risikogebiete der Frühsommer-Meningoenzephalitis (FSME) in Deutschland. *Epid Bull.* 2021, 9:3-20 (in German)