Epidemiology of *Trichinella* infection in wild boar from Spain and its impact on human health during the period 2006–2019

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**ARTICLE INFO**

**Keywords:**
Trichinellosis
Trichinella
Wild boar
Prevalence
Zoonosis
Hunting

**ABSTRACT**

Trichinellosis is a notifiable zoonotic disease caused by parasitic nematode larvae belonging to the genus *Trichinella*. Domestic pig and wild boar are important hosts within the natural cycle of *T. spiralis*, the last one being an animal whose populations have experienced an important growth. Therefore, this paper studies the prevalence of *Trichinella* infection in wild boar in Spain, as well as its relation with hunting and its impact on public health during the period 2006–2019. For this purpose, different sources of information were consulted and analyzed depending on the autonomous communities of Spain and years. During the fourteen years of study, the number of wild boars hunted and the number of cases of *Trichinella* infection in them increased (from 172 cases in 2006 to 421 in 2019), although prevalence values remained low as the number of animals analyzed also increased. On the other hand, trichinellosis in humans tended to decrease (from a peak of 107 cases in 2007 to 11 cases in 2019). Nevertheless, the numbers of both wild boars and humans infected with *Trichinella* in Spain are among the highest in Europe, and this emphasizes the importance of food safety, sanitary controls of game meat and citizen awareness campaigns, which prevent the spread of *Trichinella* through the human population.

1. Introduction

Trichinellosis is a zoonotic disease caused by consuming meat infected with *Trichinella* spp. larvae (Pozio, 2015). This is a genus of parasitic nematodes including ten species and three different genotypes (Pozio and Zarlenga, 2005, 2013; Pozio and Murrell, 2006; Pozio, 2007; Krivokapich et al., 2012; Zarlenga et al., 2020; Marucci et al., 2021). At the global level, the main source of human trichinellosis is pork from domestic pigs, followed by meat from wild boars, wild carnivores and other domestic animals (e.g., dog, horse) (Pozio, 2015; Rostami et al., 2017). In Europe, four *Trichinella* species (*T. spiralis*, *T. nativa*, *T. britovi* and *T. pseudospiralis*) are endemic in domestic and wild animals (Pozio, 1998), with *T. spiralis* and *T. britovi* being the most relevant ones due to their distribution and impact on human health (Pozio et al., 2009), although *Trichinella spiralis* causes more serious pathologies and has a higher associated mortality rate (Pozio and Murrell, 2006). In the European Union, the incidence of human trichinellosis has shown a decreasing trend during the last decades (EFSA, 2007; Murrell and Pozio, 2011; European Food Safety Authority, 2019), although in industrialized countries cases of trichinellosis due to the consumption of meat of wild boar and other game animals have increased up to several hundred cases (Rostami et al., 2017; European Food Safety Authority, 2019).

In Spain, trichinellosis is an endemic and notifiable disease, managed by the National Epidemiological Surveillance Network (RENAVE for its Spanish acronym), which follows the European regulations (European Commission, 2015). This control is of concern because trichinellosis is a threat not only for public health, but also for the economy, especially for the stockbreeding and hunting sectors (Gottstein et al., 2009). During the period 2007–2010, Spain exceeded the European average in terms of prevalence of *Trichinella* infection in wild boar (Cárdenas Contreras, 2012). Therefore, it is not surprising that most cases of trichinellosis in humans are caused by consumption of wild boar meat (Rodríguez de las Parras et al., 2004; Cárdenas Contreras, 2012; Escobar et al., 2019). In this country, *Trichinella spiralis*, *T. pseudospiralis* and *T. britovi* are involved in trichinellosis epidemiology (Pérez-Pérez et al., 2019), and mixed infection by *T. spiralis* and *T. britovi* in a wild boar was reported from Cáceres (Extremadura, central Spain) (Rodríguez et al., 2008). With regards to human trichinellosis, up to now only descriptions of sporadic outbreaks (Rodríguez-Osorio et al., 1999; Gómez-Garcia et al.,...
Table 1
Wild boar hunting bag in six autonomous communities of Spain during the period 2006–2019.

| Year | Catalonia | Andalusia | Aragon | Castile-Leon | Castile-La Mancha | Extremadura | Total | Spain |
|------|-----------|-----------|--------|--------------|-------------------|------------|-------|-------|
| 2006-09 | 24418 | 27549 | 25264 | 17955 | 31680 | 13662 | 145165 | 149221 |
| 2007-08 | 23789 | 30710 | 26612 | 16796 | 35194 | 14258 | 147359 | 160422 |
| 2008-09 | 24976 | 31407 | 26451 | 16273 | 26247 | 13299 | 138653 | 115950 |
| 2009-10 | 22238 | 35145 | 29595 | 18145 | 34606 | 14873 | 154872 | 136356 |
| 2010-11 | 25247 | 37297 | 28237 | 17918 | 30770 | 15485 | 165334 | 161061 |
| 2011-12 | 29696 | 38908 | 30166 | 22314 | 38621 | 15725 | 175280 | 209357 |
| 2012-13 | 32665 | 40358 | 36242 | 22479 | 36134 | 17535 | 185413 | 222692 |
| 2013-14 | 35393 | 42581 | 35378 | 24994 | 41304 | 17952 | 197602 | 268655 |
| 2014-15 | 33579 | 38879 | 38319 | 30764 | 41631 | 19392 | 202646 | 274728 |
| 2015-16 | 36447 | 38649 | 38117 | 29793 | 50301 | 20564 | 213871 | 310280 |
| 2016-17 | 48547 | 46724 | 40993 | 30643 | 57859 | 22700 | 247466 | 354648 |
| 2017-18 | 57090 | 51089 | 46842 | 39847 | 66168 | 24319 | 285355 | 370770 |
| 2018-19 | 65351 | 51938 | 49885 | 41229 | 66857 | 24319 | 299579 | 373225 |
| Average | 36438 | 40072 | 35680 | 26528 | 44592 | 19079 | 202272 | 239070 |

Table 2
Hunting licenses in six autonomous communities of Spain from 2006 to 2019.

| Year | Catalonia | Andalusia | Aragon | Castile-Leon | Castile-La Mancha | Extremadura | Total | Spain |
|------|-----------|-----------|--------|--------------|-------------------|------------|-------|-------|
| 2006  | 80091     | 308016    | 47022  | 140681       | 139438          | 75664      | 790852 | 983321 |
| 2007  | 80094     | 301171    | 46412  | 139984       | 149455          | 75657     | 755677 | 898036 |
| 2008  | 68360     | 288065    | 53439  | 129170       | 91915           | 73875     | 704824 | 916421 |
| 2009  | 77712     | 266554    | 52816  | 126634       | 145034          | 78822     | 747572 | 1032242|
| 2010  | 74476     | 289874    | 52553  | 124198       | 161994          | 80472     | 802572 | 1095191|
| 2011  | 65173     | 275362    | 44100  | 123462       | 128732          | 61180     | 697952 | 906347 |
| 2012  | 68828     | 257054    | 51321  | 122690       | 101050          | 68036     | 668979 | 848243 |
| 2013  | 59812     | 252779    | 49375  | 101620       | 89382           | 54461     | 607429 | 851894 |
| 2014  | 53860     | 250087    | 50814  | 101403       | 103439          | 50846     | 610449 | 825373 |
| 2015  | 47837     | 254161    | 50947  | 96111        | 104677          | 42813     | 596546 | 826777 |
| 2016  | 42087     | 252255    | 50044  | 90827        | 106406          | 40684     | 582303 | 827776 |
| 2017  | 41081     | 244886    | 45393  | 89172        | 107908          | 34506     | 562946 | 769551 |
| 2018  | 37481     | 235094    | 44958  | 89131        | 105090          | 32364     | 544118 | 860678 |
| 2019  | 35112     | 223690    | 47296  | 87229        | 96360           | 16234     | 505921 | 860957 |
| Average | 59429     | 262071    | 49035  | 111594       | 116491          | 53691     | 648476 | 886939 |

Table 3
Occurrence of Trichinella spp. in wild boar in six autonomous communities of Spain. The third column is the percentage of samples analyzed with respect to the wild boars hunted.

| Year | Catalonia | Andalusia | Aragon | Castile-Leon | Castile-La Mancha | Extremadura | Total | Spain |
|------|-----------|-----------|--------|--------------|-------------------|------------|-------|-------|
| 2008-09 | 4547     | 10        | 15.36  | 0.22         | 0.08              | 0.26       |
| 2009-10 | 8028     | 14        | 27.84  | 0.17         | 0.12              | 0.20       |
| 2010-11 | 6381     | 15        | 12.79  | 0.24         | 0.04              | 0.05       |
| 2011-12 | 3295     | 4         | 26.74  | 0.12         | 0.02              | 0.05       |

| Year | Castile-La Mancha | Extremadura | Samples | Positives | Percentage | Prevalence | Samples | Positives | Percentage | Prevalence |
|------|-------------------|-------------|---------|-----------|------------|-----------|---------|-----------|------------|-----------|
| 2008-09 | 15158          | 21         | 43.80   | 0.14      | 0.08       | 0.30     |
| 2009-10 | 12979          | 33         | 32.64   | 0.25      | 0.18       | 0.31     |
| 2010-11 | 39701          | 114        | 59.38   | 0.29      | 0.19       | 0.68     |
| 2011-12 | 25546          | 142        | 44.11   | 0.56      | 0.30       | 0.50     |
Generally, such sporadic familiar outbreaks occur within the wild boar game and domestic pig slaughtering seasons, and are associated with the consumption of meat products without sanitary control (Pérez-Pérez et al., 2019). In Germany, *T. spiralis* was detected in meat products imported from Spain (Pozio et al., 2000). Also, an important outbreak of human trichinellosis related to the consumption of wild boar meat imported from Spain was recently reported (Messiaen et al., 2016).

Research focused on the wild boar (*Sus scrofa*, Linnaeus 1758) as one of the main reservoirs of *T. spiralis* gains importance due to its demographic growth for more than a decade (Moral Moral et al., 2017). This fact, together with the practice of one type of hunting that promotes overcrowding of some animals, could favor the increase of the prevalence of certain diseases (e.g., African swine fever, brucellosis, Aujeszky’s disease) (Martínez Pulido, 2014; Martínez Pulido et al., 2018).

The main objective of this research is to update the epidemiology of *Trichinella* infection in wild boar in Spain during the years 2006–2019 and to analyze its relation with the number of animals hunted each season and with human cases of trichinellosis during the same period, in order to assess its impact on public health.

2. Material and methods

2.1. Data collection

For this study hunting bag data, such as the number of wild boars hunted and the total hunting licenses, were collected from 2006 to 2019, both at national and regional levels. These data were obtained from the Annual Directory of Forest Statistics of the Ministry of Agriculture, Fisheries and Food of the Spanish Government, the Environment Departments of each region (autonomous communities) and a bibliographical screening at the Web of Science.

Additionally, official reports of the European Food Safety Authority (EFSA) from the *EFSA Journal*, the annual epidemiological reports of the European Centre for Disease Prevention and Control (ECDC) and the Office International des Epizooties (OIE) reference laboratory reports activities were also consulted to gather information about the number of samples analyzed and the number of positive cases of *Trichinella* infection in wild boar in Spain and other European countries. In the same way, annual reports of the National Epidemiological Surveillance Network were also consulted to address the epidemiology of trichinellosis in humans, also from 2006 to 2019 and both at national and regional levels.

2.2. Estimation of trichinellosis prevalence and incidence

Prevalence of *Trichinella* infection in wild boar was calculated as the percentage of infected hosts with respect to the total number of hosts examined (Margolis et al., 1982; Bush et al., 1997). Nevertheless, we were not able to estimate incidence of the disease in this host species, since accurate information about wild boar abundance was not available. Regarding human trichinellosis, we also estimated the incidence as the number of cases/100000 persons and year.

2.3. Statistical analysis

From the data obtained in the first section of Material and methods, a comparison was made between different autonomous communities to determine which ones were most affected by *Trichinella* in recent years, both in wild boars and in humans. The autonomous communities...
analyzed for this study were: Andalusia, Extremadura, Castile-La Mancha, Castile and Leon, Aragon and Catalonia. The remaining Spanish autonomous communities were discarded due to a lack of information. Furthermore, the available hunting bag data made it possible to verify whether the presence of trichinellosis was higher in the regions with more hunting activity. With this aim, we applied Pearson’s correlation analyses between the hunted wild boars (whose value is corrected according to the prevalence: that is HxP, where H is the number of hunted wild boars and P is the prevalence) and the positive cases of trichinellosis in humans within each autonomous community and year.

We used generalized linear mixed-effects models (GLMM) and generalized additive mixed models (GAMM) to assess the association between the presence of Trichinella larvae in wild boar muscles and the total number of wild boars hunted (explanatory variables) on the incidence of the trichinellosis in humans (dependent variable). We considered the variable “year” as a random factor in the model; this variable indicates the year in which data were collected, and the GLMM was conducted with a Gaussian distribution. We used a GAMM model with the aim of testing whether linearity can really be assumed in the model, to examine the possibility that the effect of the explanatory variables on the response variable had an unknown non-linear form. Thus, in order to gain flexibility we must select the most appropriate functional form in

**Table 4**

Cases of trichinellosis in humans in six autonomous communities of Spain from 2006 to 2019. Incidence is expressed as the number of cases/100000 inhabitants and year. The number in red is the average of Spain incidence.

| Year   | Catalonia | Andalusia | Aragon | Castile-Leon | Castile-La Mancha | Extremadura | Total | Spain | Incidence (Spain) |
|--------|-----------|-----------|--------|--------------|-------------------|-------------|-------|-------|-------------------|
| 2005-06| 4         | 2         | 24     | 0            | 0                 | 0           | 26    | 36    | 0.081             |
| 2006-07| 0         | 59        | 13     | 34           | 0                 | 0           | 106   | 107   | 0.237             |
| 2007-08| 0         | 1         | 1      | 4            | 0                 | 7           | 23    | 25    | 0.110             |
| 2008-09| 0         | 0         | 7      | 9            | 0                 | 3           | 11    | 0     | 0.053             |
| 2009-10| 0         | 0         | 0      | 3            | 0                 | 7           | 23    | 17    | 0.036             |
| 2010-11| 0         | 0         | 0      | 16           | 0                 | 0           | 30    | 0     | 0.064             |
| 2011-12| 0         | 0         | 2      | 21           | 0                 | 0           | 23    | 23    | 0.049             |
| 2012-13| 0         | 0         | 0      | 0            | 0                 | 1           | 28    | 0     | 0.059             |
| 2013-14| 0         | 0         | 0      | 0            | 1                 | 0           | 1     | 1     | 0.002             |
| 2014-15| 0         | 0         | 0      | 2            | 1                 | 1           | 3     | 3     | 0.006             |
| 2015-16| 0         | 13        | 0      | 0            | 0                 | 0           | 13    | 14    | 0.030             |
| 2016-17| 2         | 0         | 2      | 0            | 0                 | 0           | 4     | 5     | 0.011             |
| 2017-18| 1         | 0         | 0      | 0            | 0                 | 0           | 1     | 2     | 0.004             |
| 2018-19| 1         | 0         | 0      | 0            | 0                 | 0           | 1     | 1     | 0.023             |
| Total  | 7         | 74        | 41     | 78           | 34                | 7           | 237   | 353   | 0.055             |

**Fig. 2.** Occurrence of *Trichinella* infection in wild boar in Spain from 2006 to 2019.
of wild boars hunted and the prevalence of the Trichinella infection in wild boar

Fig. 3. Three-dimensional plots for the interaction effects of the total number of wild boars hunted and the prevalence of the Trichinella infection in wild boar on incidence of trichinellosis in humans.

Table 5
Summary of the significant estimates for GLMM and GAMM models. The explanatory variables are the prevalence of the Trichinella infection in wild boar and the number of total wild boars hunted, and the dependent variable is the incidence of trichinellosis in humans.

| Model         | R² adj | Parameters     | Coefficients | p-value |
|---------------|--------|----------------|--------------|---------|
| GLMM          | 0.6136 | Intercept      | -2.22754     | 0.014   |
|               |        | Hunting        | -0.00001     | 0.0008  |
|               |        | Prevalence     | 8.87673      | 0.0275  |
| GAMM          | 0.589  | Intercept      | 2.66e-09     |         |
|               |        | Hunting        | 0.0004       |         |
|               |        | Prevalence     | 0.0185       |         |

We also compared the incidence of trichinellosis in humans among the European Union (EU) countries with the highest incidence values. The data analyzed are interannual values for each country, that is, we have an annual entry value of the incidence in humans for each country and year of study, which make up a total of 14 homogeneous entries for each of the 10 countries considered in the study. To carry out this comparison, we used the Kruskal-Wallis test instead of the more common ANOVA due to the lack of normality in the data. This nonparametric test was used to perform a comparison between the distribution of the different groups in order to detect significant differences between countries. Multiple comparisons after the Kruskal-Wallis test were carried out using Dunn’s test.

We used R software 4.0.5 (R Development Core Team, 2021) to conduct all the statistical analyses. Package alme (Pinheiro et al., 2017) was used to fit the GLMM model, with the lme function. Package mgcv (Wood, 2017) was used to fit the GAMM model, using the gamm function. For an appropriate fitting of the GLMM and GAMM models in R, we have followed the recommendations given by Zuur et al. (2009). For nonparametric multiple comparisons, we used the dunn.test function to conduct the Kruskal-Wallis test using stats package, and the post-hoc Dunn’s tests were conducted using the package dunnTest. All the statistical graphs were produced via the ggplot function in R package ggplot2 (Wickham, 2016).

3. Results

3.1. Wild boar hunting bag in Spain

The number of hunted wild boars provided by the autonomous communities studied represents, on average, 84.8% of the number of hunted for the whole country (Table 1). Castile-La Mancha is the autonomous community with the highest average of hunted wild boars in the fourteen years of study, reaching quantities of more than 66,000 specimens during the 2016-17 and 2017-18 seasons. This region is closely followed by Catalonia with a peak of more than 65,000 wild boars, Andalusia with nearly 52,000 and Aragon with approximately 50,000 specimens, all of them hunted during the 2017-18 season. As can be seen in Table 1, the number of hunted wild boars has increased significantly in the autonomous communities analyzed. However, the number of hunting licenses (and, therefore, the number of hunters) presents an opposite trend, as shown in Table 2.

3.2. Cases of Trichinella infection in wild boar

As can be seen in Table 3, the number of wild boar samples analyzed has increased considerably in just ten years. This greater number of samples has been also paralleled by significant increase in the number of Trichinella cases in several autonomous communities, such as Catalonia, Andalusia, Castile-La Mancha and Extremadura. Even so, the prevalence values decreased in some regions such as Catalonia and Aragon (Fig. 1). The increase in the number of cases of Trichinella infection in wild boar can be also observed at national level, just as Fig. 2 shows.

3.3. Cases of human trichinellosis

The number of cases of human trichinellosis contributed by the autonomous communities analyzed represents, on average, 67.1% of the number of cases for the whole country. Data evidenced a peak (>100 cases) in the season 2006-2007, followed by a decreasing trend (Table 4).

3.4. Statistical results

In all regions or autonomous communities studied, the number of cases of Trichinella infection in wild boar did not correlate with the
number of cases of trichinellosis in humans, since all p-values were more than 0.05 (from 0.1556 in Castile and Leon to 0.9656 in Extremadura).

We found significant differences for the parameters of the two variables considered, i.e. the prevalence of the *Trichinella* infection in wild boar and the number of total wild boars hunted, for the two models GLMM and GAMM. Table 5 shows the outputs of the two models: as can we see, all the p-values are less than the significant level, thereby, our models are significant. For both models, the prevalence of the *Trichinella* infection in wild boar shows a positive influence on the incidence of the trichinellosis in humans, and the number of total wild boars hunted shows a negative influence on the same incidence (Table 5). GLMM model explained 61.36% of the variance of the incidence, and the GAMM model explained 58.9% of the variance of the incidence (see Table 5). GLMM and GAMM models provided similar results. The goodness of fit tests of the two models are included in the Supplementary Material (Figs. S1 and S2).

On the other hand, Fig. 3 shows the linear relations of the explanatory variables on the response in a tridimensional plane. In this figure, we appreciate graphically how the number of wild boars hunted and the prevalence of the *Trichinella* infection affect the incidence in humans.

Finally, Kruskal-Wallis test shows (chi-squared = 87.571, df = 9, p-value = 4.994e-15) a p-value lower than 5%; therefore, we reject the null hypothesis that the samples come from the same population. As we can see in Table 6, there are significant differences among the incidence values of the European countries considered. Fig. 4 shows the results of the multiple pairwise comparisons of Dunn’s test graphically: there are no significant differences between the incidence in Bulgaria and Romania; on the other hand, neither we found significant differences between the incidence in Latvia and Lithuania, nor between France and Germany, nor Italy and Poland.

4. Discussion

Wild boar populations in Spain have increased in recent years, as can be seen in Table 1. This increase would be positively conditioned by the decreasing number of hunters, represented by the hunting licenses registered each season (Table 2). In addition, the absence of large predators and the abandonment of rural areas also affect this population growth, together with the enormous adaptability of the wild boar, which can tolerate human pressure and live in varied territories, including those close to urban centers (Ballesteros, 1998; Mitchell-Jones et al., 1999; Azorit and Moro, 2010).

The population growth of wild boar, together with a greater concern on the part of the government, have also caused a general increase in the number of samples analyzed to detect *Trichinella* and, therefore, an increase in the number of cases of *Trichinella* infection detected (Table 3). In other *Trichinella* models, density-dependent mechanisms were highlighted Mikkonen et al. (2005) found a positive correlation between rat population density and *Trichinella* spp. prevalence, and Airas et al. (2010) reported a similar trend between *Trichinella* spp. prevalence and the density of raccoon dogs (*Nyctereutes procyonoides*) in Finland. Nevertheless, the growing number of wild boar is apparently not paralleled by a higher prevalence of *Trichinella* infection in this particular host; however, caution is needed, since wild boar sampling was locally suboptimal. Ultimately, the prevalence of *Trichinella* infection in wild boar may reflect two important factors in the epidemiology of this group of parasites: the presence of *Trichinella britovi*, a carnivore-adapted species, whose larvae survive only a few weeks/months in the muscles of swine (Pozio et al., 2020), and the reduction of the scavenger activity of reservoir animals (González et al., 2021), i.e. the main form of *Trichinella* transmission, when abundant anthropogenic food resources are available. Anyway, it is difficult to explain the trend of the
Trichinella spp. prevalence in wild boar in Spain because of the lack of information about the Trichinella species involved in the reported cases, and its transmission pathways between wildlife-wild boar and domestic pigs.

On the other hand, all cases of trichinellosis detected in humans (Table 4) were associated with the consumption of wild boar meat infected with Trichinella, originated in hunts without any type of sanitary control. However, infection of farmed wild boars or pigs has been practically non-existent, since very few cases of trichinellosis have been detected in them in recent years. Therefore, the presence of trichinellosis in humans in Spain consists of sporadic cases (outbreaks) in which Trichinella spiralis and T. britovi infections since both species show differences regarding pathogenicity in humans and infectivity/persistence in wild boars (Pozio et al., 2009).

Finally, the analysis of Trichinella infection in Europe places Spain as the European country with the second highest number of cases in wild boar, only behind Poland; although, in prevalence levels, Bulgaria tops the list. On the other hand, Spain is the European country with the fourth highest number of cases of trichinellosis in humans, while Romania is the first. The data are included in the Supplementary Material (Tables S1 and S2).

5. Conclusions

Just as previous research demonstrated (Moral Moral, 2016; Moral et al., 2017), the hunting data analyzed for this study shows that wild boar populations continue rising. On the contrary, the number of hunters tends to decrease, which undoubtedly may have helped the population growth of the species.

On the other hand, cases of Trichinella infection in wild boar have also increased both at the national level and in some autonomous communities of Spain, although the prevalence values remain low. However, an opposite trend has been observed in humans: although there have been some important outbreaks during the fourteen years of study, the cases of trichinellosis in humans tend to decrease through the years. This result demonstrates the importance of food safety programs, since most of the positive cases involve people who had consumed wild boar meat without any type of sanitary control.

Finally, the analysis of Trichinella in Europe places Spain as the European country with the second highest number of cases in wild boar and the fourth highest number of cases in humans.

Declaration of competing interest

All authors drafted the manuscript and revised it for final approval.

Acknowledgements

The authors are indebted to Dr PG Meneguz (Torino University) for kindly providing data on trichinellosis in Italy. The research activities of the authors are partially supported by the Junta de Andalucía (RNM-118 and RNM-175 groups).

Appendix A. Supplementary data

Supplementary data to this article can be found at https://doi.org/10.1016/j.ijppaw.2022.07.008.

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