

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

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Abstract

Trichinellosis is a notifiable zoonotic disease caused by parasitic nematode larvae belonging to genus *Trichinella*. Domestic pig and wild boar are important hosts within the natural cycle of *T. spiralis*, being the last one 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 bag and its impact on public health during the period 2006-2019. For this purpose, different sources of information were consulted and analyzed depending on 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 tends to decrease (from a peak of 107 cases in 2007 to 11 cases in 2019). Nevertheless, both the number of 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 *Trichinella* spreads through the human population.

Keywords: trichinellosis, *Trichinella*, wild boar, prevalence, zoonosis, hunting

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; Pozio and Murrell, 2006; Pozio, 2007; Krivokapich *et al.*, 2012; Pozio and Zarlenga, 2013; 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), being *T. spiralis* and *T. britovi* 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 last decades (EFSA, 2007; Murrell and Pozio, 2011; EFSA and ECDC, 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; EFSA and ECDC, 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 not only is a threat for the 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-García *et al.*, 2003) or regional studies (Pérez Pérez *et al.*, 2019) have been reported. Generally, such sporadic familiar outbreaks occur within the wild boar game season and house domestic pig slaughtering, and are associated to 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 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 promote overcrowding of some animals, could favour 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 Épizooties (OIE) reference laboratory reports activities were also consulted to gather information about 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 one were the most affected by *Trichinella* in recent years, both in wild boar 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 casted aside due to a lack of information. Furthermore, the available hunting bag data made 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 $H \times P$, 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 treat the possibility that the effect of the explanatory variables on the response variable had an unknown non-linear form. Thus, to gain flexibility, we must select the most appropriate functional form in our model.

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 Kruskal-Wallis test instead of 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 Kruskal-Wallis test were carried out using the Dunn's test.

We used R software 4.0.5 (R Development Core Team 2021) to conduct all the statistical analyses. Package *nlme* (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 *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 *kruskal.test* function to conduct Kruskal-Wallis test using *stats* package, and the post-hoc Dunn's test were conducted using the package *dunnTest*. The whole statistical graphs were produced via *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, the 84.8 % of the number of hunted for the whole country (Table 1). Castile-La Mancha is the autonomous community with higher 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 also caused a 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 (Figure 1). The increase in the number of cases of *Trichinella* infection in wild boar can be also observed at national level, just as Figure 2 shows.

3.3. Cases of human trichinellosis

The number of cases of human trichinellosis contributed by the autonomous communities analyzed represents, on average, the 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 was not correlated 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 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 test of the two models are included in the Supplementary Material (Figures S1 and S2).

Finally, Figure 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 are affecting on the incidence in humans.

4. Discussion

Wild boar populations in Spain have increased in recent years, such 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 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, 1999; Azorit and Moro, 2010).

The population growth of wild boar, together with a greater concern 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 fact, the more population density, the more overcrowding and ease of transmission of diseases and parasites. Mikkonen et al. (2005) found positive correlation between rat population density and *Trichinella* spp. prevalence, and Airas et al. (2020) reported a similar trend between *Trichinella* spp. prevalence and density of raccoon dogs (*Nyctereutes procyonoides*) in Finland. Unfortunately data on wild boar density are scarce. Nevertheless, prevalence remains low and relatively constant; however, it does not represent the actual wild boar populations, due to the percentage of samples analyzed is very low in some autonomous communities. Ultimately, the prevalence of *Trichinella* infection in wild boar probably reflects two important factors in the epidemiology of this group of parasites: the presence of *Trichinella britovi*, whose larvae survive a few weeks/months in the muscles of swine, and the strong reduction of the scavenger activity of reservoir animals (González *et al.*, 2021), i.e. the main way of *Trichinella* transmission, due to the increased availability of anthropogenic food resources. Anyway, it is difficult to explaining the trend of the *Trichinella* spp. prevalence in wild boar in Spain because 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 barely 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 people relax preventive measures or show excessive trust towards other people who give away or sell hunting products. This is proved by Pearson's correlation analyses, which do not show a statistically significant relation between the number of wild boars hunted and the cases of trichinellosis in humans. Furthermore, the GLMM accounted for 61.36 % of the variance of the incidence of trichinellosis in humans, while the GAMM explained 58.9 %. This means that, in addition to the number of wild boars harvested and prevalence of trichinellosis in this species, other factors may also affect epidemiology of human trichinellosis (e.g., sylvatic transmission between other wild host species, sociocultural, technical (regarding protocols for processing game meat or subproducts) and, even, legislative). Anyway, our study was limited, as available data do not separate between *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 second European country with the 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 fourth European country with the highest number of cases of trichinellosis in humans, while Romania is the first one. The data are included in the Supplementary Material (Tables S1 and S2).

5. Conclusions

Just as previous research demonstrated (Moral Moral, 2016; Moral 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 tends to decrease throughout 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 second European country with the highest number of cases in wild boar and the fourth with the highest number of cases in humans.

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References

- Airas, N., Saari, S., Mikkonen, T., Virtala, A.M., Pellikka, J., Oksanen, A., Isomursu, M., Kilpela, S.S., Lim, C.W., Sukura, A. 2010. Sylvatic *Trichinella* spp. infection in Finland. *J Parasitol.* 96, 67-76. Doi: 10.1645/GE-2202.1
- Azorit, C., Moro, J., 2010. El jabalí (*Sus scrofa* Linnaeus, 1758). In: Santiago Moreno, J. & López Sebastián, A. (coords) Ungulados silvestres de España: biología y tecnologías reproductivas para su conservación y aprovechamiento cinegético. INIA, Madrid, pp 215-240.
- Ballesteros, F., 1998. Las especies de caza en España: biología, ecología y conservación. Estudio y Gestión del Medio, Oviedo. <https://dialnet.unirioja.es/servlet/libro?codigo=137902>
- Bush, A.O., Lafferty, K.D., Lotz, J.M., Shostak, A.W., 1997. Parasitology meets ecology on its own terms: Margolis *et al.* revisited. *J. Parasitol.* 83, 575–583. <https://doi.org/10.2307/3284227>
- Cárdenas Contreras, Z.L., 2012. La vigilancia de triquina en España: situación actual y valoración de un sistema alternativo. Master Dissertation, Autonomous University of Barcelona, Barcelona.
- Escobar, C.F., Navarro, A.R., García, O.D., Sánchez, E.M., 2019. Vigilancia epidemiológica de brotes de triquinosis en España. Temporadas 2006/07 a 2013/14. *Bol. Epidemiol. Sem.* 27, 32-38. <http://revista.isciii.es/index.php/bes/article/view/1089>
- European Commission. 2015. Commission Implementing Regulation (EU) 2015/1375 of 10 August 2015 laying down specific rules on official controls for *Trichinella* in meat (Text with EEA relevance). http://data.europa.eu/eli/reg_impl/2015/1375/oj
- European Food Safety Authority (EFSA), 2007. The community summary report on trends and sources of zoonoses, zoonotic agents, antimicrobial resistance and foodborne outbreaks in the European Union in 2006. *EFSA J.* 130, 197–206. <https://doi.org/10.2903/j.efsa.2007.130r>
- European Food Safety Authority (EFSA), European Centre for Disease Prevention and Control (ECDC), 2019. The European Union One Health 2018 Zoonoses Report. *EFSA J.* 17, 5926. <https://doi.org/10.2903/j.efsa.2019.5926>

- Gómez-García, V., Hernández-Quero, J., Rodríguez-Osorio, M., 2003. Short report: human infection with *Trichinella britovi* in Granada, Spain. Am. J. Trop. Med. Hyg. 68, 463-464. <https://doi.org/10.4269/ajtmh.2003.68.463>
- González, M., Martínez-Carrasco, C., Sánchez-Zapata, J.A., Moleón, M., 2021. Smart carnivores think twice: Red fox delays scavenging on conspecific carcasses to reduce parasite risk. Appl. Anim. Behav. Sci. 243, 105462. <https://doi.org/10.1016/j.applanim.2021.105462>
- Gottstein, B., Pozio, E., Nöckler, K., 2009. Epidemiology, diagnosis, treatment and control of trichinellosis. Clin. Microbiol. Rev. 22, 127–145. <https://doi.org/10.1128/CMR.00026-08>
- Krivokapich, S.J., Pozio, E., Gatti, G.M., Prous, C.L.G., Ribicich, M., Marucci, G., La Rosa, G., Confalonieri, V., 2012. *Trichinella patagoniensis* n. sp. (Nematoda), a new encapsulated species infecting carnivorous mammals in South America. Int. J. Parasitol. 42, 903–910. <https://doi.org/10.1016/j.ijpara.2012.07.009>
- Margolis, L., Esch, G.W., Holmes, J.C., Kuris, A.M., Schad, G., 1982. The use of ecological terms in parasitology (report of an *ad hoc* committee of the American Society of Parasitologists). J. Parasitol. 68, 131-133. <https://doi.org/10.2307/3281335>
- Martínez Pulido, M.A., 2014. Seroprevalencia de *Brucella* spp, enfermedad de Aujeszky, peste porcina clásica, peste porcina africana y enfermedad vesicular porcina en jabalíes cazados en diferentes comarcas de la provincia de Jaén. Master Dissertation, University of Córdoba, Córdoba.
- Martínez Pulido, M.A., Azorit, C., Arenas Casas, A., 2018. Resultados de la vigilancia epidemiológica del jabalí en cinco comarcas agrarias de la provincia de Jaén durante la temporada cinegética 2013-14. Comunicación al II Congreso Nacional de Sanidad Animal, Córdoba.
- Marucci, G., Romano, A.C., Interisano, M., Toce, M., Pietragalla, I., Collazzo, G.P., Palazzo, L., 2021. *Trichinella pseudospiralis* in a red kite (*Milvus milvus*) from Italy. Parasitol. Res. 120, 2287–2290. <https://doi.org/10.1007/s00436-021-07165-0>
- Messiaen, P., Forier, A., Vanderschueren, S., Theunissen, C., Nijs, J., Van Esbroeck, M., Bottieau, E., De Schrijver, K., Gyssens, I.C., Cartuyvels, R., Dorny, P., van der Hilst, J., Blockmans, D., 2016. Outbreak of trichinellosis related to eating imported wild boar meat, Belgium, 2014. Eurosurveillance. 21, 30341. <https://doi.org/10.2807/1560-7917.ES.2016.21.37.30341>
- Mikkonen, T., Valkama, J., Wihlman, H., Sukura, A. 2005. Spatial variation of *Trichinella* prevalence in rats in Finnish waste disposal sites. J. Parasitol. 91, 210-213. Doi: 10.1645/GE-3230RN

- Mitchell-Jones, A.J., Amori, G., Bogdanowicz, W., Krystufek, B., Reijnders, P.J.H., Spitzenberger, F., Stubbe, M., Thissen J.B.M., Vohralík, V., Zima, J., 1999. The atlas of European mammals. T & AD Poyser, London.
<https://www.nhbs.com/the-atlas-of-european-mammals-book>
- Moral Moral, S., 2016. Análisis de la situación de las poblaciones de jabalí (*Sus scrofa*) en España mediante el estudio de antecedentes bibliográficos, datos oficiales de caza y animales abatidos en cacerías. Bs Degree Dissertation, Jaén University, Jaén.
- Moral Moral, S., Carrasco, R., López Montoya, A.J., Azorit, C., 2017. Índices de caza a partir de registros oficiales como herramienta para evaluar la evolución de las poblaciones de jabalí en España. Ann. R. Acad. C. Vet. And. Or. 30, 39-54. <https://dialnet.unirioja.es/servlet/articulo?codigo=7402809>
- Murrell, K.D., Pozio, E., 2011. Worldwide occurrence and impact of human trichinellosis, 1986-2009. Emerg. Infect. Dis. 17, 2194–2202. <https://doi.org/10.3201/eid1712.110896>
- Pérez-Pérez, A., Guimbao Bescós, J., Cebollada Gracia, A.D., Malo Aznar, C., Martínez Cuenca, S., Aznar Brieba, A., Lázaro Belanche, M.A., Sanz Lacambra, I., Compés Dea, C., 2019. Brotes epidémicos de triquinosis ocurridos en Aragón durante el período 1998-2017. Rev. Esp. Salud Pública. 93, e201902005. https://scielo.isciii.es/scielo.php?script=sci_arttext&pid=S1135-57272019000100101
- Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D., Heisterkamp, S., Van Willigen, B., Maintainer, R., 2017. Package ‘nlme’. Linear and nonlinear mixed effects models, version, 3(1).
- Pozio, E., 1998. Trichinellosis in the European Union: epidemiology, ecology and economic impact. Parasitol. Today. 14, 35-38. [https://doi.org/10.1016/S0169-4758\(97\)01165-4](https://doi.org/10.1016/S0169-4758(97)01165-4)
- Pozio, E., 2007. Taxonomy, biology and epidemiology of *Trichinella* parasites. (ed. Dupouy-Camet, J., Murrell, K.D.) FAO/WHO/OIE Guidelines for the surveillance, management, prevention and control of trichinellosis. 1-35. <http://www.fao.org/3/a0227e/a0227e.pdf>
- Pozio, E., 2015. *Trichinella* spp. imported with live animals and meat. Vet. Parasitol. 213, 46–55. <https://doi.org/10.1016/j.vetpar.2015.02.017>
- Pozio, E., Murrell, K.D., 2006. Systematics and epidemiology of *Trichinella*. Adv. Parasitol. 63, 367–439. [https://doi.org/10.1016/S0065-308X\(06\)63005-4](https://doi.org/10.1016/S0065-308X(06)63005-4)
- Pozio, E., Nöckler, K., Hoffman, L., Voigt, W.P., 2000. Autochthonous and imported *Trichinella* isolates in Germany. Vet. Parasitol. 87, 157-161. [https://doi.org/10.1016/s0304-4017\(99\)00179-x](https://doi.org/10.1016/s0304-4017(99)00179-x)

- Pozio, E., Rinaldi, L., Marucci, G., Musella, V., Galati, F., Cringoli, G., Boireau, P., La Rosa, G., 2009. Hosts and habitats of *Trichinella spiralis* and *Trichinella britovi* in Europe. *Int. J. Parasitol.* 39, 71–79. <https://doi.org/10.1016/j.ijpara.2008.06.006>
- Pozio, E., Zarlenga, D.S., 2005. Recent advances on the taxonomy, systematics and epidemiology of *Trichinella*. *Int. J. Parasitol.* 35, 1191–1204. <https://doi.org/10.1016/j.ijpara.2005.07.012>
- Pozio, E., Zarlenga, D.S., 2013. New pieces of the *Trichinella* puzzle. *Int. J. Parasitol.* 43, 983–997. <https://doi.org/10.1016/j.ijpara.2013.05.010>
- Rodríguez de las Parras, E., Rodríguez Ferrer, M., Nieto Martínez, J., Ubeira, F.M., Gárate Ormaechea, T., 2004. Revisión de los brotes de triquinelosis detectados en España durante 1990-2001. *Enf. Inf. Microbiol. Clin.* 22, 70–76. <https://doi.org/10.1157/13056885>
- Rodríguez, E., Olmedo, J., Ubeira, F.M., Blanco, C., Gárate, T., 2008. Mixed infection, *Trichinella spiralis* and *Trichinella britovi*, in a wild boar hunted in the Province of Cáceres (Spain). *Exp. Parasitol.* 119, 430-432. <https://doi.org/10.1016/j.exppara.2008.03.017>
- Rodríguez-Osorio, M., Abad, J.M., de Haro, T., Villa-Real, R., Gómez-García, V., 1999. Human trichinellosis in southern Spain: serologic and epidemiologic study. *Am. J. Trop. Med. Hyg.* 61, 834-837. <https://doi.org/10.4269/ajtmh.1999.61.834>
- Rostami, A., Gamble, H.R., Dupouy-Camet, J., Khazan, H., Bruschi, F., 2017. Meat sources of infection for outbreaks of human trichinellosis. *Food Microbiol.* 64, 65–71. <https://doi.org/10.1016/j.fm.2016.12.012>
- Wickham, H., 2016. *ggplot2: Elegant Graphics for Data Analysis*. New York: Springer-Verlag. <https://link.springer.com/book/10.1007%2F978-0-387-98141-3>
- Wood, S.N., 2017. *Generalized Additive Models: An Introduction with R* (2nd edition). Chapman and Hall/CRC Press. <https://doi.org/10.1201/9781315370279>
- Zarlenga, D., Thompson, P., Pozio, E., 2020. *Trichinella* species and genotypes. *Res. Vet. Sci.* 133, 289-296. <https://doi.org/10.1016/j.rvsc.2020.08.012>
- Zuur, A.F., Ieno, E.N., Walker, N.J., Saveliev, A.A., Smith, G.M., 2009. *Mixed Effects Models and Extensions in Ecology with R*. Springer. <https://link.springer.com/book/10.1007/978-0-387-87458-6#toc>

Figure legends

Figure 1. Prevalence of *Trichinella* infection in wild boar from several Spanish autonomous communities.

Figure 2. Occurrence of *Trichinella* infection in wild boar in Spain since 2006 to 2019.

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

Table 1. Wild boar hunting bag in six autonomous communities of Spain during the period 2006 - 2019.

WILD BOAR HUNTING BAG								
	Catalonia	Andalusia	Aragon	Castile-Leon	Castile-La Mancha	Extremadura	Total	Spain
2005-06	24418	27549	25264	17955	31680	13662	145165	149221
2006-07	23789	30710	26612	16796	35194	14258	147359	160422
2007-08	24976	31407	26451	16273	26247	13299	138653	115950
2008-09	22238	35415	29595	18145	34606	14873	154872	136356
2009-10	25947	37397	28837	17918	39770	15485	165354	161601
2010-11	29696	38908	30016	22314	38621	15725	175280	209357
2011-12	32665	40358	36242	22479	36134	17535	185413	222692
2012-13	35393	42581	35378	24994	41304	17952	197602	268655
2013-14	33579	38879	38319	30764	41631	19392	202564	274728
2014-15	36447	38649	38117	29793	50301	20564	213871	310280
2015-16	48547	46724	40993	30643	57859	22700	247466	354648
2016-17	57090	51089	46842	39847	66168	24319	285355	370770
2017-18	65351	51938	49885	41229	66857	24319	299579	373225
2018-19	50000	49409	46974	42242	57920	33027	279572	
AVERAGE	36438	40072	35680	26528	44592	19079	202722	239070

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

HUNTING LICENSES								
	Catalonia	Andalusia	Aragon	Castile and Leon	Castile-La Mancha	Extremadura	Total	Spain
2006	80091	308016	47022	140681	139438	75604	790852	983321
2007	80094	271171	46412	139984	149455	68561	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		703095	957191
2011	65173	275302	44100	123465	128732	61180	697952	906437
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	
2019	35112	223690	47296	87229	96360	16234	505921	
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 hunted wild boars.

CATALONIA				
	Samples	Positives	Percentage	Prevalence
2008-09	295	6	1.33	2.03
2009-10	282	6	1.09	2.13
2017-18	8662	44	13.25	0.51
2018-19	13371	31	26.74	0.23

ANDALUSIA				
	Samples	Positives	Percentage	Prevalence
2008-09	19263	6	54.39	0.03
2009-10	21006	17	56.17	0.08
2017-18	51938	94	100	0.18
2018-19				

ARAGON				
	Samples	Positives	Percentage	Prevalence
2008-09	4547	10	15.36	0.22
2009-10	8028	14	27.84	0.17
2017-18	6381	15	12.79	0.24
2018-19	3295	4		0.12

CASTILE-LEON				
	Samples	Positives	Percentage	Prevalence
2008-09	5794	15	31.93	0.26
2009-10	5887	7	32.86	0.12
2017-18	15583	7	37.80	0.04
2018-19	17328	51	41.02	0.29

CASTILE-LA MANCHA				
	Samples	Positives	Percentage	Prevalence
2008-09	15158	21	43.80	0.14
2009-10	12979	33	32.64	0.25
2017-18	39701	114	59.38	0.29
2018-19	25546	142	44.11	0.56

EXTREMADURA				
	Samples	Positives	Percentage	Prevalence
2008-09	12478	38	83.90	0.30
2009-10	15485	28	100	0.18
2017-18	24319	165	100	0.68
2018-19	33027	176	100	0.53

Table 4. Cases of trichinellosis in humans in six autonomous communities of Spain since 2006 to 2019. Incidence is expressed as the number of cases/100000 inhabitants and year. Number in red is average of Spain incidence.

CASES OF TRICHINELLOSIS IN HUMANS									
	Catalonia	Andalusia	Aragon	Castile-Leon	Castile-La Mancha	Extremadura	Total	Spain	Incidence (Spain)
2005-06	4		2	24			26	36	0.081
2006-07	0	59	13	34	0	0	106	107	0.237
2007-08	0	1	1	4			6	51	0.110
2008-09	0	0	7	9	0	7	23	25	0.053
2009-10	0	0	0	3	11	0	14	17	0.036
2010-11	0	0	16	0	0	0	16	30	0.064
2011-12	0	0	0	2	21	0	23	23	0.049
2012-13	0	0	0	0	0	0	0	28	0.059
2013-14	0	0	0	0	1	0	1	1	0.002
2014-15	0	0	0	2	1	0	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					1	11	0.023
TOTAL	7	74	41	78	34	7	237	353	0.055

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 the trichinellosis in humans.

Model	R^2 -adj	Parameters	Coefficients	p-value
GLMM	0.6136	Intercept	-2.22754	0.0141
		Hunting	-0.00001	0.0008
		Prevalence	8.87673	0.0275
GAMM	0.589	Intercept		3.66e-09
		Hunting		0.0004
		Prevalence		0.0185