

REVIEW

Global prevalence of *Trichinella* in pigs: A systematic review and meta-analysis

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Abstract

Background: Investigating the global epidemiological patterns of *Trichinella* in pigs is required for accurate recognition and to establishing proper control programmes and preventive measures, as well as to decrease human exposure.

Objectives: To obtain a better understanding of the global prevalence of *Trichinella* in domestic pigs and factors that might influence the prevalence, a systematic review and meta-analysis was performed.

Methods: The Preferred Reporting Items for Systematic Reviews and Meta-Analyses guidelines were followed. Multiple databases were used to identify literature published between January 2000 and December 2021, representing studies from 1985 to 2021, on *Trichinella* prevalence in domestic pigs. Prevalence was calculated on a global and country level, by country Human Development Index (HDI), climate, pig management system, and diagnostic test.

Results: The global pooled prevalence based on 60 manuscripts representing 32 countries and 65 pig populations was 2.02% (95% confidence interval [CI]: 0.88–3.62) and the estimated pooled prevalence in different continents ranged from 0.00% to 11.8%. *Trichinella* was highest in low HDI countries (21.6%; 95% CI: 4.3–47.2), tropical wet

Aida Vafae Eslahi and Meysam Olfatifar contributed equally to this study.

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climates (20.9%; 95% CI: 10.3–34.1), and non-intensive pig farming systems (6.1%; 95% CI: 1.0–15.5).

Conclusions: While there were regional differences in *Trichinella* prevalence in domestic pigs, these were likely driven by country HDI and pig farming systems versus climatic factors. Increased biosecurity in outdoor pig production systems, focused meat inspections and promoting awareness could potentially decrease risk of infection.

KEYWORDS

epidemiology, prevalence, swine, trichinellosis

1 | INTRODUCTION

Foodborne pathogens continue to be a serious health and economic concern in developed and developing countries (Sekamatte et al., 2018; Zhao et al., 2014; Badri, Olfatifar, KarimiPourSaryazdi et al., 2022). A broad range of protozoan and helminthic parasites are responsible for foodborne diseases, some of which are zoonotic with the potential to be fatal for humans (e.g., *Trichinella* spp., *Taenia solium*) (Murrell, 2013). *Trichinella* spp. are ubiquitous foodborne helminths with sylvatic or domestic cycles infecting humans and a wide variety of mammals including pigs, horses, rats, foxes, bears, and seals (Devleeschauwer et al., 2015; Dupouy-Camet, 2000; Pozio, 2000). Omnivores and carnivores with predator, scavenger, and cannibalistic habits are the main reservoirs for *Trichinella* spp. (Feidas et al., 2014; Maleki et al., 2020). All of the 13 taxa (nine species and four genotypes) of the parasite are pathogenic for humans (Gómez-Morales et al., 2018; Mukaratirwa et al., 2013; Sharma et al., 2020). *Trichinella* was first observed by James Paget, who found the parasite in specimens he obtained from a cadaver in 1835. However, Richard Owen formalized this report as a publication, and between the 1850s and 1870s Rudolf Virchow and Zenker determined the life cycle and pathogenicity for humans (Campbell, 1983; Devleeschauwer et al., 2015; Schultz, 2008).

Trichinellosis (also called trichinosis), the disease caused by *Trichinella* spp., has been reported in humans from 55 countries and has been observed to infect domestic and/or wild animals worldwide, except Antarctica (Devleeschauwer et al., 2015; Dupouy-Camet, 2000; Feidas et al., 2014). It is a disease of public health importance with non-specific manifestations varying from fever, abdominal pain, diarrhoea, nausea, vomiting, to severe lesions which are associated with myalgia, myocarditis, and encephalitis due to larval migration (Rawla & Sharma, 2021; Sun et al., 2018; Wang et al., 2020). Most human trichinellosis cases are caused by migrating larvae of *Trichinella spiralis*, which is associated with domestic pigs (Murrell, 2013; Rawla & Sharma, 2021; Wang et al., 2020); hence, *T. spiralis* infection in humans is predominantly limited to the regions of the world where pork is widely consumed although in some geographic locations, due to culture, horse meat and dog meat can contribute to outbreaks (Rostami et al., 2017). Humans, as accidental hosts, primarily acquire the infection by consumption of raw or inadequately cooked pork and pork-derived products containing infective larvae in muscle, although infection through the consumption of wild game (boar and

bear) is increasing in some regions (Barruet et al., 2020; Bilska-Zajac et al., 2020; Diaz et al., 2020; Murrell, 2013; Rostami et al., 2017). Domesticated pigs become infected via the consumption of uncooked meat or carcasses of other animals that are infected, including rats and via tail biting of other pigs that are infected. A considerable economic loss in pork production is attributable to the infection since it usually remains undetected and untreated in the live animal (Wang et al., 2020; Murrell, 2013).

After consumption of infected muscle, the larvae are released during gastric digestion and pass through four molts in the intestinal epithelium. They then mature into adult parasites; the males die after copulation while the gravid females penetrate the intestinal mucosa and lay pre-larvae. The larvae migrate and become located in the striated muscles, organs (e.g., heart and lungs) and central nervous system (namely the brain) where they develop into a fully developed first stage larva (L1) and encyst inside a typical lemon-shaped cyst (except for *Trichinella pseudospiralis*, *Trichinella papuae*, and *Trichinella zimbabwensis*, which develop also to L1, but without cyst) (Dupouy-Camet, 2000; Wang et al., 2020).

The geographic distribution of *Trichinella* is affected by the survival of larvae in the muscle tissue of decaying host carcasses, cultural eating habits, and interventions in domestic and wild habitats (Feidas et al., 2014). *Trichinella* parasites are capable of performing an anaerobic metabolism to increase their survival time in decomposing tissues. The length of survival of larvae in muscle tissue indicates the probability of the carcasses being consumed by scavenger hosts (Rossi et al., 2019). Survival time is highly related to the size of the host's body, since the decomposition site in micromammals is more affected by environmental conditions of temperature and humidity (Pozio, 2000).

Prevention of *Trichinella* in pigs is achieved through management approaches including grain feeding or cooking of any fed refuse, indoor production, and ensuring that rat carcasses on premises are removed. Freezing, cooking, and irradiation are the recommended procedures for inactivating *Trichinella* larvae in pork to prevent transmission to humans. In some countries, farms can be certified *Trichinella* free, and in other countries there is testing of pork at slaughter with this latter being more common. All routine diagnostic methods for *Trichinella* are based on the direct finding of larvae in muscle. *Trichinella* larvae predilection sites can differ based on species. In pigs, the diaphragm pillars, tongue, and masseter muscles can be collected for detection of larvae. However, using diaphragm tissue has an advantage as it can

be digested easily (Gajadhar et al., 2019). Three common methods of finding larvae in muscle are as follows: the squash (compression) method in which muscle is compressed between slides and examined microscopically, trichoscopy in which muscle samples are magnified and projected onto a screen, and pooled sample digestion in which an enzyme and acid are used to digest the meat releasing the larvae (Dupouy-Camet, 2006). The digestion method is practical, reliable, and cheap and has become the preferred method for food safety purposes, including routine slaughter inspection. It is more sensitive than trichoscopy and is efficient, especially in non-endemic regions (Forbes et al., 2003). Among the various digestion methods in use (e.g., the stomacher method and Trichomatic 351), the magnetic stirrer method is the most widely recognized and is recommended by various authorities as the gold standard. The level of sensitivity of these methods is influenced by the muscle sample examined, the amount of sample, the enzyme used in digestion, and the quality assurance of the related method (Gajadhar et al., 2019). In addition to these methods, PCR can be used to identify infections and differentiate *Trichinella* species, although it is used primarily as a research and surveillance tool and not employed in meat inspection (Bilska-Zajac et al., 2022; Zarlenga et al., 1999). Antibody and antigen tests (e.g., ELISA and western blot) also can be used with some demonstrating exposure and others being more specific for an active infection (Braasch et al., 2020; Pozio et al., 2020). As with PCR, these are used in epidemiological studies and not in meat inspection.

Even with the implementation of preventive strategies for *Trichinella* in pigs and inspection of meat for human consumption, the disease remains a hazard in most countries (Murrell, 2013; Pozio, 2014). Investigating the global epidemiological patterns of *Trichinella* in pigs is required for accurate recognition, as well as establishing proper control programmes and preventive measures. The current review and meta-analysis evaluates available scientific reports on the prevalence of *Trichinella* in pigs with the purpose of estimating the global pooled prevalence and studying the associated risk factors.

2 | MATERIALS AND METHODS

2.1 | Search methodology and inclusion and exclusion criteria

A systematic review and meta-analysis based on the Preferred Reporting Items for Systematic Reviews and Meta-Analyses guidelines (<http://www.prisma-statement.org/>) was performed. Multiple databases (Web of Science, PubMed, ProQuest, Scopus, and Google Scholar) were searched for literature on *Trichinella* in domestic pigs. Keywords, used alone or in combination, were as follows: Trichinellosis, *Trichinella spiralis*, *Trichinella britovi*, *Trichinella pseudospiralis*, *Trichinella nelsoni*, *Trichinella* spp., Food borne parasite, Pig, *Sus domesticus*, Swine, Pork, Piglet, Piggy, Prevalence, Epidemiology, Frequency, Worldwide, Global (Supporting Information 1). The titles and abstracts were screened, and duplicates and irrelevant records were excluded. Two independent authors evaluated the full texts of the remaining articles.

The references of the full-text articles were reviewed to determine whether any potentially applicable articles had not been identified through the database search.

The inclusion criteria were as follows: (1) peer-reviewed articles containing original data; (2) published prior to December 21, 2021 and after January 1, 2000; (3) in English, Spanish, or Portuguese; (4) cross-sectional studies evaluating the prevalence of *Trichinella* in domestic pigs in some region of the world; (5) accessible abstract and full-text article; and (6) numerator and denominator data available to confirm prevalence. Articles were excluded if they did not meet the above criteria, including review articles with no original data, letters, editorials, and articles with confusing/undetermined results. Articles that focused on testing specifically due to an outbreak (in people or pigs) in the region also were excluded.

2.2 | Data extracted

A Microsoft® Excel® 2016 MSO (16.0.4498.1000) spreadsheet was prepared to extract the following data from the included articles: first author's name, country where the study was conducted, continent, year of publication, genus and species, animal management system (various, non-intensive, intensive), and diagnostic method as primary factors and sample (meat/muscle or serum) and muscle type, as secondary data. In addition, based on the location of the study, the following data were added to the Excel® spreadsheet as primary data for analysis: Human Development Index (HDI; <https://hdr.undp.org/en/composite/HDI>), climate (<https://www.britannica.com/science/Koppen-climate-classification>), and average temperature and rainfall (<https://en.climate-data.org/>); country income level (<https://datahelpdesk.worldbank.org/knowledgebase/articles/906519-world-bank-country-and-lendinggroups>), annual precipitation (<https://en.climate-data.org/>), and humidity (<https://www.timeanddate.com/weather/>) were included as secondary data for analysis (Eslahi, Olfatifar, et al., 2022).

2.3 | Study quality assessment

A Newcastle-Ottawa Scale for cross-sectional studies was employed to evaluate the quality of the included studies (Modesti et al., 2016; Badri, Olfatifar, Karim et al., 2022). Briefly, the scores were assigned based on the three following domains: selection (maximum of five stars), comparability (maximum of two stars), and outcome (maximum of three stars) (Eslahi, Hashemipour et al., 2022; Badri, Olfatifar, Wandra et al., 2022; Mirzadeh et al., 2021).

2.4 | Data synthesis and statistical analysis

To assess consistency of the results, and to establish methods considering the combination of the included studies, the heterogeneity between studies was measured using a Cochrane's Q test and the I^2 statistic.

The I^2 values of <25%, 25%–75%, >75% were categorized as low, moderate, and high heterogeneity, respectively. A p -value less than 0.05 was regarded as significant heterogeneity (Higgins et al., 2003). The random-effects model was applied to estimate the pooled prevalence. The Freeman–Tukey Double Arcsine Transformation was used to stabilize the variances, prior to the pooling of data (Doi & Xu, 2021). Pooled prevalence was also calculated for the primary subgroups; diagnostic method, species of *Trichinella*, climate, average temperature, annual rainfall, and animal management and the secondary subgroups sample type, HDI, annual precipitation, humidity, and muscle type. Chi-square test was used to investigate differences within subgroups. A meta-regression analysis was performed to indicate the impact of average temperature, and year of publication on the prevalence. Egger’s funnel plot and Begg’s funnel plot as well as Doi plot were implemented to indicate the possible publication bias (Furuya-Kanamori et al., 2020). All statistical analyses were carried out using the meta-package of R (version 3.6.1) (R Core Team, 2020).

3 | RESULTS

A total of 9610 articles were initially identified during the database searches, including 416 from PubMed, 948 from Scopus, 51 from ProQuest, 725 from Web of Science, and 7470 from Google Scholar (Figure 1 and Supporting Information 1). After excluding duplicates and applying inclusion/exclusion criteria, studies were assessed at the full text level, of which 60 studies (comprising 751,167,472 animals) were included in the systematic review and meta-analysis (Figure 1, Table 1

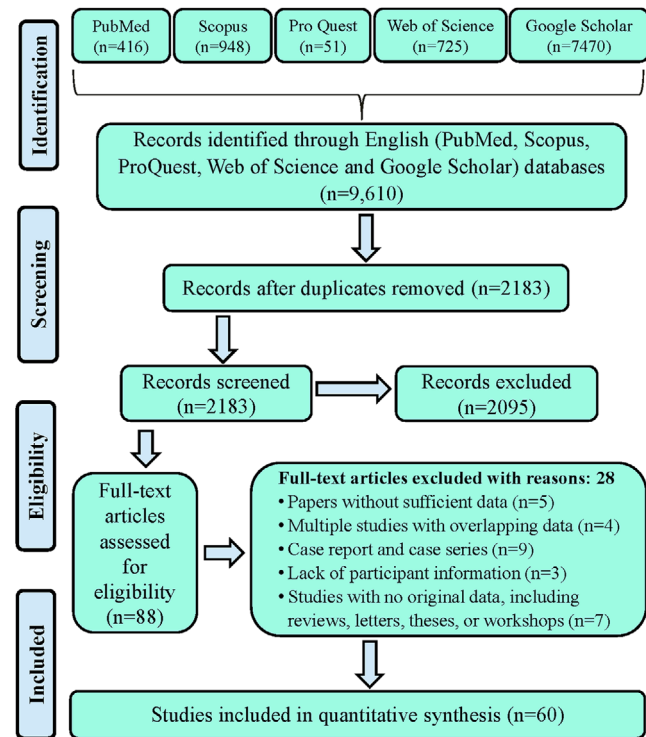


FIGURE 1 Flow diagram of the study design process

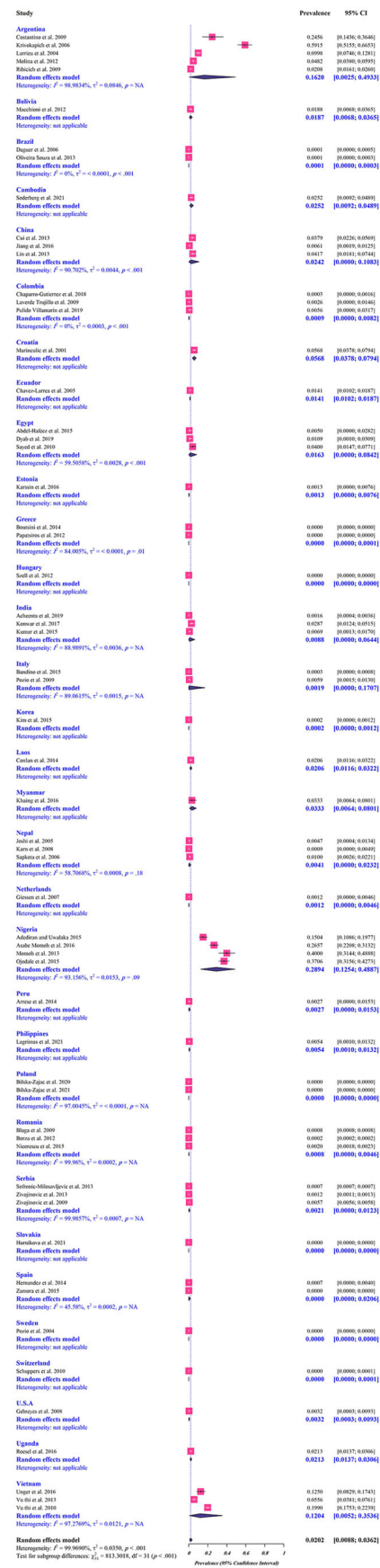


FIGURE 2 Forest plots for random-effects meta-analysis of *Trichinella* in pigs based on countries

TABLE 1 Main characteristics of eligible studies reporting *Trichinella* in pigs

No.	Author	Year [†]	Study Years	Continent	Sample size	Number Positive	Country	Sample type	Genus and species
1	Marinculic et al.	2001	NS	Europe	475	27	Croatia	Serum	<i>Trichinella</i> spp.
2	Larrieu et al.	2004	2000–2002	South America	481	48	Argentina	Meat/serum	<i>Trichinella</i> spp.
3	Pozio et al.	2004	1985–2003	Europe	66,000,000	27	Sweden	Meat	<i>Trichinella</i> spp.
4	Chavez-Larrea et al.	2005	2000–2003	South America	2977	42	Ecuador	Serum	<i>Trichinella</i> spp.
5	Joshi et al.	2005	NS	Asia	425	2	Nepal	Serum	<i>Trichinella spiralis</i>
6	Daguer et al.	2006	2004–2005	South America	6264	0	Brazil	Meat	<i>Trichinella</i> spp.
7	Krivokapich et al.	2006	1996–2005	South America	164	97	Argentina	Meat	<i>Trichinella spiralis</i>
8	Sapkota et al.	2006	2004–2005	Asia	400	4	Nepal	Serum	<i>Trichinella</i> spp.
9	Giessen et al.	2007	NS	Europe	845	1	Netherlands	Serum	<i>Trichinella spiralis</i>
10	Gebreyes et al.	2008	NS	North America	616	2	USA	Serum	<i>Trichinella spiralis</i>
11	Karn et al.	2008	2006–2007	Asia	576	0	Nepal	Meat	<i>Trichinella</i> spp.
12	Blaga et al.	2009	1997–2004	Europe	34,540,315	27928	Romania	Meat	<i>Trichinella</i> spp.
13	Costantino et al.	2009	NS	South America	57	14	Argentina	Serum	<i>Trichinella</i> spp.
14	Laverde Trujillo et al.	2009	NS	South America	194	0	Colombia	Meat	<i>Trichinella</i> spp.
15	Pozio et al.	2009	2006	Europe	681	4	Italy	Meat	<i>Trichinella</i> spp.
16	Ribicich et al.	2009	NS	South America	3224	67	Argentina	Meat/serum	<i>Trichinella spiralis</i>
17	Zivojinovic et al.	2009	1995–2006	Europe	1,554,262	8889	Serbia	Meat	<i>Trichinella</i> spp.
18	Sayed et al.	2010	2006–2007	Africa	150	6	Egypt	Meat	<i>Trichinella</i> spp.
19	Schuppers et al.	2010	2006–2007	Europe	20164	0	Switzerland	Meat	<i>Trichinella</i> spp.
20	Vu Thi et al.	2010	NS	Asia	1035	206	Vietnam	Serum	<i>Trichinella spiralis</i>
21	Borza et al.	2012	1998–2011	Europe	6,195,756	1088	Romania	Meat	<i>Trichinella</i> spp.
22	Macchioni et al.	2012	2007 & 2011	South America	320	6	Bolivia	Meat/serum	<i>Trichinella</i> spp.
23	Molina et al.	2012	2001–2010	South America	1516	73	Argentina	Meat/serum	<i>Trichinella</i> spp.
24	Papatsiros et al.	2012	2009–2010	Europe	2,121,460	7	Greece	Meat	<i>Trichinella britovi</i> /spp.
25	Szell et al.	2012	2006–2011	Europe	16,000,000	0	Hungary	Meat	<i>Trichinella</i> spp.

(Continues)

TABLE 1 (Continued)

No.	Author	Year [†]	Study Years	Continent	Sample size	Number Positive	Country	Sample type	Genus and species
26	Cui et al.	2013	2010–2011	Asia	475	18	China	Meat	<i>Trichinella</i> spp.
27	Lin et al.	2013	NS	Asia	192	8	China	Meat	<i>Trichinella</i> spp.
28	Momoh et al.	2013	2011	Africa	120	48	Nigeria	Serum	<i>Trichinella</i> spp.
29	de Oliveira Souza et al.	2013	2009–2011	South America	9520	0	Brazil	Meat	<i>Trichinella spiralis</i>
30	Sofronic-Milosavljevic et al.	2013	2001–2010	Europe	21,616,000	15312	Serbia	Meat	<i>Trichinella spiralis</i>
31	Vu Thi et al.	2013	NS	Asia	558	31	Vietnam	Serum	<i>Trichinella</i> spp.
32	Zivojinovic et al.	2013	2009–2010	Europe	282,960	344	Serbia	Meat	<i>Trichinella spiralis</i>
33	Arrese et al.	2014	2011–2012	South America	185	0	Peru	Meat/seru	<i>Trichinella</i> spp.
34	Boutsini et al.	2014	2009–2012	Europe	4,534,889	37	Greece	Meat	<i>Trichinella</i> spp.
35	Conlan et al.	2014	2008–2009	Asia	728	15	Laos	Meat	<i>Trichinella britovi</i> /spp.
36	Hernández et al.	2014	2008–2009	Europe	709	0	Spain	Serum	<i>Trichinella</i> spp.
37	Abdel-Hafeez et al.	2015	2014–2015	Africa	100	0	Egypt	Meat	<i>Trichinella</i> spp.
38	Adediran and Uwalaka	2012	2010	Africa	246	37	Nigeria	Serum	<i>Trichinella</i> spp.
39	Bandino et al.	2015	2010–2014	Europe	7585	2	Italy	Meat	<i>Trichinella britovi</i>
40	Kim et al.	2015	2013	Asia	2350	0	Korea	Serum	<i>Trichinella</i> spp.
41	Kumar et al.	2015	2011	Asia	432	3	India	Meat	<i>Trichinella spiralis</i>
42	Nicorescu et al.	2015	2014	Europe	113,383	227	Romania	Meat	<i>Trichinella</i> spp.
43	Ojodale et al.	2015	2013	Africa	286	106	Nigeria	Serum	<i>Trichinella spiralis</i>
44	Zamora et al.	2015	2006–2014	Europe	314,853,949	384	Spain	Meat	<i>Trichinella</i> spp.
45	Momoh et al.	2016	NS	Africa	350	93	Nigeria	Serum	<i>Trichinella</i> spp.
46	Jiang et al.	2016	2014–2015	Asia	823	5	China	Meat	<i>Trichinella spiralis</i>
47	Kärssin et al.	2016	2012	Europe	374	0	Estonia	Serum	<i>Trichinella</i> spp.
48	Khaing et al.	2016	2012	Asia	90	3	Myanmar	Meat	<i>Trichinella</i> spp.
49	Roesel et al.	2016	2013–2015	Africa	1125	24	Uganda	Serum	<i>Trichinella</i> spp.
50	Unger et al.	2016	NS	Asia	200	25	Vietnam	Serum	<i>Trichinella</i> spp.

(Continues)

TABLE 1 (Continued)

No.	Author	Year [†]	Study Years	Continent	Sample size	Number Positive	Country	Sample type	Genus and species
51	Konwar et al.	2017	NS	Asia	279	8	India	Serum	<i>Trichinella</i> spp.
52	Chaparro-Gutiérrez et al.	2018	2014–2016	South America	1773	0	Colombia	Meat/serum	<i>Trichinella</i> spp.
53	Acheenta et al.	2019	2016–2017	Asia	2445	4	India	Meat/serum	<i>Trichinella</i> spp.
54	Dyab et al.	2019	2018–2019	Africa	184	2	Egypt	Meat	<i>Trichinella spiralis</i>
55	Pulido-Villamarín et al.	2019	NS	South America	89	0	Colombia	Serum	<i>Trichinella</i> spp.
56	Bilska-Zajac et al.	2020	2009–2016	Europe	86,989,313	150	Poland	Meat	<i>Trichinella spiralis/britovi</i>
57	Bilska-Zajac et al.	2021	2012–2021	Europe	194,449,146	172	Poland	Meat	<i>Trichinella spiralis/britovi</i>
58	Hurníková et al.	2021	2007–2018	Europe	1,843,464	0	Slovakia	Meat	<i>Trichinella</i> spp.
59	Lagrimas et al.	2021	2017	Asia	555	3	Philippines	Serum	<i>Trichinella</i> spp.
60	Söderberg et al.	2021	2019	Asia	238	6	Cambodia	Serum	<i>Trichinella</i> spp.

Abbreviation: NS, not stated.

[†]Year of publication.**TABLE 2** Sub-group analysis of the global prevalence of *Trichinella* in pigs

Variable	Number of studies	Sample size	Number positive	Pooled prevalence % (95% CI) [†]	Heterogeneity [‡]	
					I ²	τ ²
Diagnostic method						
Digestion method	26	716,292,004	26,468	0.7 (0.0; 2.4)	99	0.028
Digestion and squash	3	292,630	350	0.5 (0.0; 10.4)	95	0.009
Digestion and ELISA	1	3224	67	2.0 (1.6; 2.6)	NA	NA
Digestion, ELISA, and Western blot	2	20740	0	0.0 (0.0; 0.4)	25	0.000
ELISA	21	14579	618	4.3 (1.1; 9.4)	98	0.045
ELISA and squash	1	185	0	0.2 (0.0; 1.5)	NA	NA
ELISA and western blot	5	3068	150	7.5 (0.0; 19.7)	96	0.028
ELISA, western blot, and immunofluorescence	1	57	14	24.5 (14.3; 36.4)	NA	NA
Squash	4	34,540,793	27,930	0.2 (0.0; 1.0)	43	0.000
Molecular (real-time PCR)	1	192	8	4.1 (1.8; 7.4)	NA	NA
Genus and species						
<i>Trichinella</i> spp.	41	469,314,060	54,178	1.9 (0.6; 3.8)	99	0.032
<i>Trichinella spiralis</i>	15	281,843,513	1248	2.6 (0.1; 8.0)	99	0.051
<i>Trichinella britovi</i>	6	288,215,776	65	0.0 (0.0; 0.0)	97	<0.000

(Continues)

TABLE 2 (Continued)

Variable	Number of studies	Sample size	Number positive	Pooled prevalence % (95% CI) [†]	Heterogeneity [‡]	
					I ²	τ ²
HDI						
Very high level	28	751,134,138	54,900	0.9 (0.0; 2.9)	99	0.035
High level	12	23367	82	0.6 (0.1; 1.5)	95	0.004
Medium level	15	7840	315	2.5 (0.8; 5.1)	97	0.013
Low level	5	2127	308	21.6 (4.3; 47.2)	98	0.047
Continent						
Asia	17	11801	341	2.3 (0.8; 4.5)	97	0.013
Europe	21	751,125,730	54,599	0.0 (0.0; 0.2)	99	0.002
Africa	8	2561	316	11.8 (1.9; 28.3)	98	0.062
North America	1	616	2	0.3 (0.0; 0.9)	NA	NA
South America	13	26,764	347	3.7 (0.1; 11.4)	98	0.061
Climate						
Hot humid continental	8	40,853,910	29,276	0.5 (0.0; 1.9)	99	0.005
Tropical savanna	6	17,395	27	0.7 (0.0; 2.8)	94	0.005
Hot desert	4	619	8	1.1 (0.0; 4.4)	63	0.002
Warm humid continental	8	365,302,936	376	0.1 (0.0; 1.0)	96	0.006
Tropical wet	7	2795	546	20.9 (10.3; 34.1)	96	0.024
Hot-summer Mediterranean	6	321,519,273	434	0.3 (0.0; 1.9)	93	0.000
Humid subtropical	11	23,460,065	24,850	4.6 (0.1; 14.7)	99	0.068
Oceanic	6	6198	49	0.5 (0.0; 1.5)	91	0.001
Hot semi-arid	3	3156	15	0.8 (0.0; 6.4)	88	0.003
Tropical rainforest	1	1125	24	2.1 (1.3; 3.0)	NA	NA
Average temperature						
>20°C	20	24,905	620	5.4 (1.9; 10.7)	99	0.042
10–20°C	33	401,839,786	54,630	1.2 (0.2; 2.9)	99	0.029
<10°C	7	349,302,781	355	0.0 (0.0; 0.4)	93	0.002
Annual rainfall						
>1500 mm	9	7454	328	3.1 (0.4; 8.1)	98	0.021
1001–1500 mm	21	49,342	645	6.2 (1.7; 13.1)	99	0.066
401–1000 mm	24	744,453,708	54,580	0.2 (0.0; 0.5)	99	0.003
<400 mm	6	6,656,968	52	0.4 (0.0; 2.0)	87	0.004
Management [§]						
Various	19	351,272,267	28,586	1.2 (0.3; 2.8)	99	0.013
Non-intensive	19	1,573,427	1040	6.1 (1.0; 15.5)	99	0.094
Intensive	8	200,045,753	863	0.1 (0.0; 0.3)	99	0.001

Abbreviation: HDI, Human Development Index.

[†]Within group data analysis was conducted using chi-square tests; within all groups there were differences ($p < 0.001$).

[‡]Heterogeneity between studies was evaluated using Cochran's Q test and the I² statistic except in cases where there was only one study (indicated as NA, not applicable). All were significant ($p < 0.001$) with the exception of ELISA and Western blot ($p = 0.25$).

[§]Management method was not indicated in all studies.

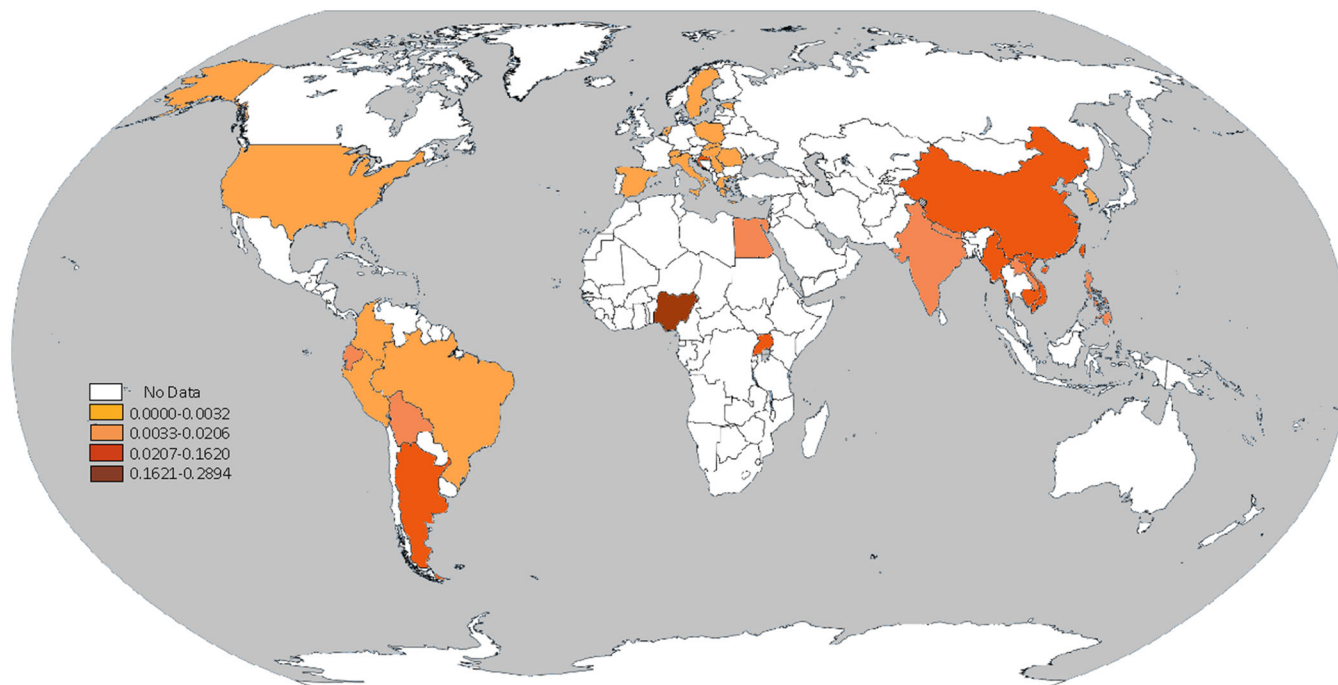


FIGURE 3 Global prevalence of *Trichinella* in pigs in different geographical regions of the world based on the included studies. White indicates an absence of studies from that country. The blank map was obtained from https://commons.wikimedia.org/wiki/File:World_Map_Blank_-_with_blue_sea.svg and edited by author.

and Supporting Information 2). Based on the Modified Newcastle-Ottawa Scale, 40 out of 60 studies had a total score of 7–9 points (high quality) and 20 had a total score of 4–6 points (moderate quality) (Table S1).

3.1 | Pooled prevalence: Global and geographical

The global pooled prevalence based on 60 manuscripts representing 32 countries and 65 pig populations was 2.02% (95% confidence interval (CI): 0.88–3.62) (Figures 2 and 3) and the estimated pooled prevalence in different continents ranged from 0.00% to 11.8% (Table 2).

3.2 | Pooled prevalence: Diagnostic method used, sample type, muscle type, and species

Several diagnostic methods were used within and across studies with digestion and ELISA being the most common. In general, the number of identified positive samples was higher in studies that used ELISA which is reflected in the results for serum samples (Table 2 and Table S2). The highest positivity rate was with the combination of ELISA, western blot, and immunofluorescence, but this combination was only used in one study. Within studies that used meat for digestion or the squash test, positivity was highest in those that used intercostal muscle and the diaphragm (8.7%, 95% CI: 0.00–1.00) (Table S2). Few studies differentiated the species of *Trichinella*; however, in those studies that did

differentiate species, prevalence was higher for *T. spiralis* 2.6% (95% CI: 0.1–8.0).

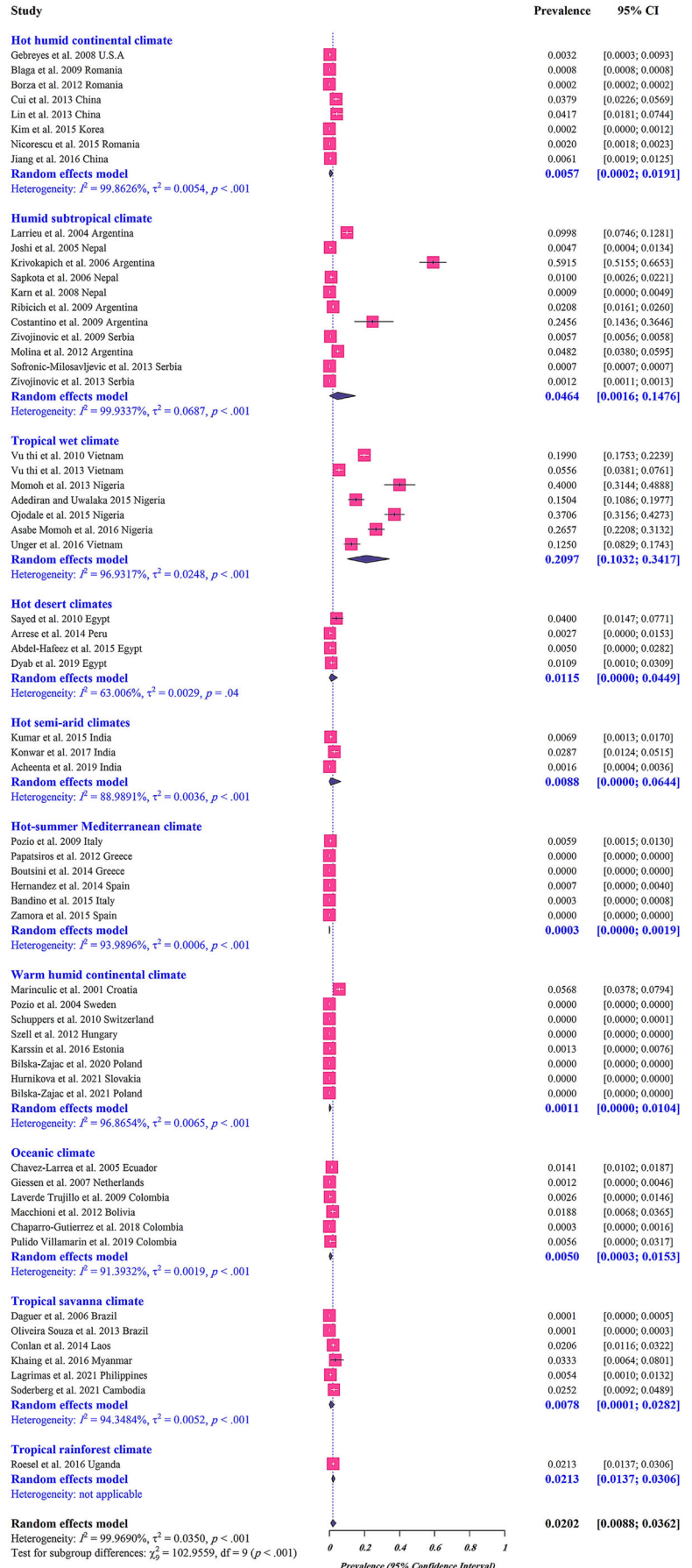
3.3 | Pooled prevalence: Climatic conditions, humidity, average temperature, annual precipitation, and annual rainfall

Concerning climate, the highest estimated pooled prevalence was in the tropical wet climate (20.9%, 95% CI: 10.3 – 34.1) (Figure 4). In the humidity range of 40%–75% (2.8%, 95% CI: 0.8 – 5.9), and the average temperature range of >20°C (5.4%, 95% CI: 1.9 – 10.7), the pooled prevalence was at its highest level (Table 2). Subgroup analysis revealed that the annual precipitation of <300 mm, and the annual rainfall of 1001–1500 mm showed the highest rate of *Trichinella* infection with a pooled prevalence of 2.5% (95% CI: 0.3 – 6.5), and 6.2% (95% CI: 1.7 – 13.1), respectively (Table 2 and Table S2).

3.4 | Pooled prevalence: Sociodemographic variables and type of animal management

Five studies fell in low level HDI countries, and these had the highest pooled prevalence (Table 2). Based on country level income, prevalence is lowest in high-income countries and highest in lower-middle income countries; however, only one study was from a low-income country (Table S2). Regarding the type of animal management, *Trichinella*

FIGURE 4 Forest plots for random-effects meta-analysis of *Trichinella* in pigs based on climatic conditions



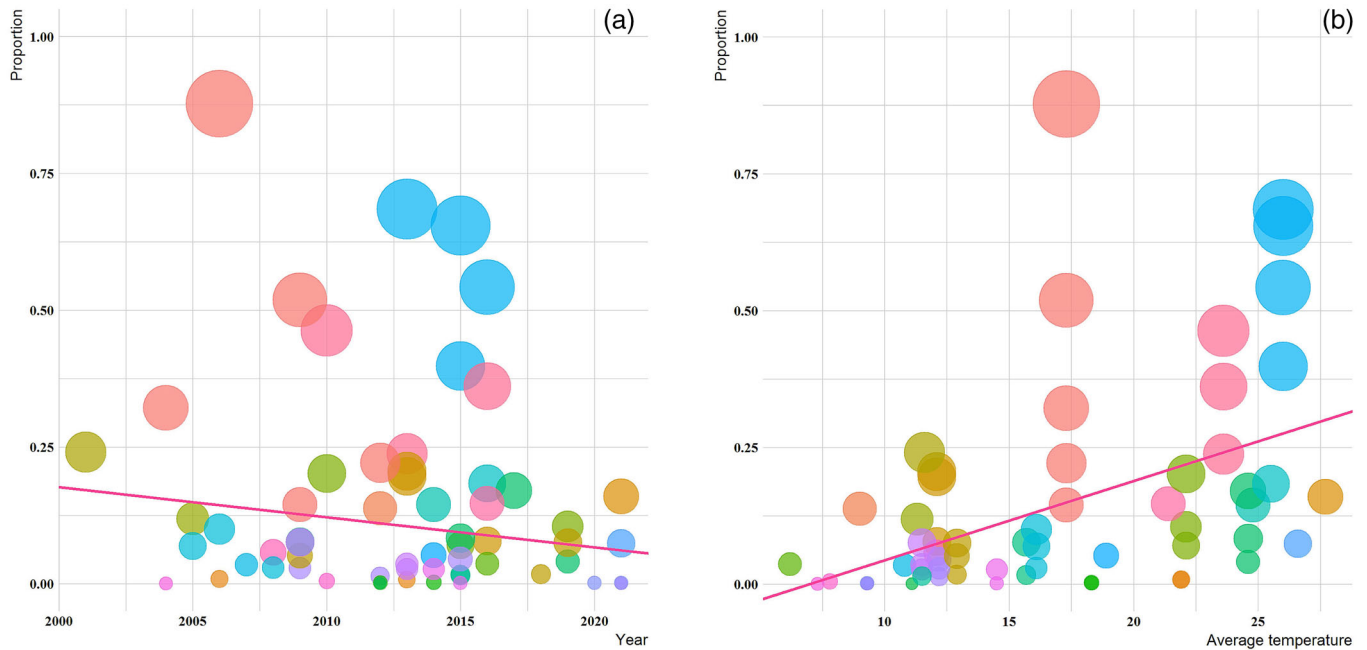


FIGURE 5 Meta-regression analysis of *Trichinella* in pigs, based on (a) year of publication, and (b) average temperature. The pink line is the regression line, which was plotted based on the intercept and the slope of the regression model. The different colour bubbles represent the countries under study and their sizes indicate the effect size of each study.

infection was the most prevalent in animals raised in a non-intensive system with a pooled prevalence of 1.6% (95% CI: 1.0 – 15.1) (Table 2).

3.5 | Meta-regression results and bias

Our analysis demonstrated that there was a significant result for the average temperature. Thus, the temperature was the source of heterogeneity (slope = -0.1014 , $p < 0.0002$). Furthermore, the relationship between the pooled prevalence and year of publication was not significant (slope = 11.1763 , $p < 0.29$) (Figure 5a,b).

A significant publication bias was detected using Egger's test ($t = 2.1611$, $p = 0.034$); however, the publication bias was not significant in Begg's test ($p = 0.068$) (Figure 6a,b). Furthermore, there was a major asymmetry in the Doi plot (LFK index: 10.37) (Figure 7).

4 | DISCUSSION

Pig meat is the primary source of *Trichinella* infection in people worldwide with boar meat and other game meat, horse meat, and dog meat contributing to outbreaks within specific regions (Rostami et al., 2017). Pig meat makes up nearly 35% of the world's meat production, 34% of global protein consumption and has an expected growth of 13% by 2030 (FAO, 2021; OECD & FAO, 2021). The increasing demand for pork meat and subsequent pig production in farms including animals living in high densities can facilitate distribution and transmission of infective pathogens (Maes et al., 2020; VanderWaal & Deen, 2018); hence, it is important to have a better understanding of the current global preva-

lence of *Trichinella* in pigs and factors that can influence prevalence. Most recent systematic reviews and meta-analyses on *Trichinella* have focused predominately on infection in people, wild boars and other wild animals and the distribution of genotypes with only regional reviews focusing on infection in domestic pigs (Devleeschauwer et al., 2015; Feidas et al., 2014; Ribicich et al., 2020; Pozio, 2019; Rostami et al., 2018). The systematic review and meta-analysis presented herein is the first to estimate the global prevalence of *Trichinella* in domestic pigs using data published in the first two decades of the 21st century. Overall, 2.0% of pigs tested positive for *Trichinella* and positivity rates appear to have decreased in at least some locations. For example, the prevalence in Greece and China is lower than what was identified in studies from the 1950s–1980s (0.02%–2.2%) and 1960s–1990s (0.0026%–27.1%), respectively (Sotiraki et al., 2001; Takahashi et al., 2000). In other locations, such as Poland, prevalence has remained low although there have been sporadic outbreaks (Bilska-Zajac et al., 2021; Ramisz et al., 2001). Our findings revealed significant geographical differences with the highest pooled prevalence in Africa, although this was heavily influenced by the studies from Nigeria which relied on ELSIAs. Given the paucity of studies from some regions, the distribution of *Trichinella* could be much wider and prevalence higher than that found in this meta-analysis, for instance, there was only one study available for North America and none for New Zealand, Papua New Guinea, and Thailand, all areas with documented infections in pigs prior to 2000 (Pozio, 2001).

The subgroup analysis suggested higher prevalence of the infection in pigs from countries with a low level HDI, suggesting that this index might be a better predictor of prevalence than country income. For example, Nigeria, with the highest prevalence, has a low level HDI but

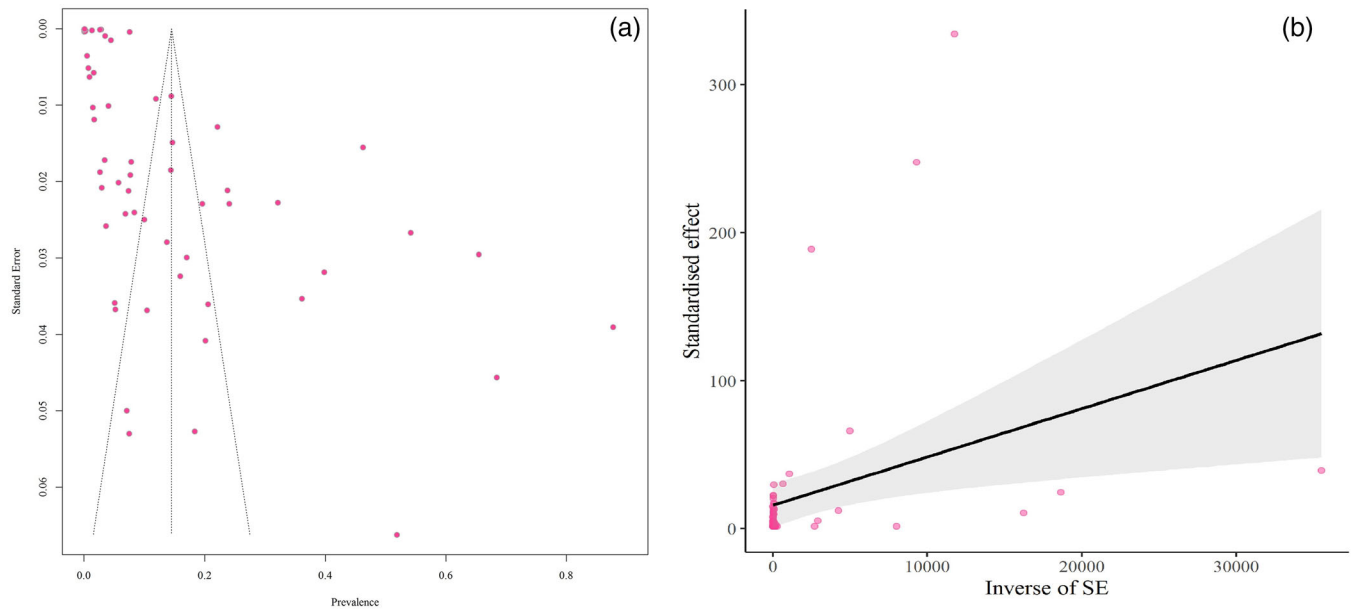


FIGURE 6 Egger's funnel plot (a) and Begg's funnel plot (b) to assess publication bias in studies evaluating *Trichinella* in pigs. Coloured circles represent each study. The middle line is the effect size and the other two lines are the corresponding confidence ranges.

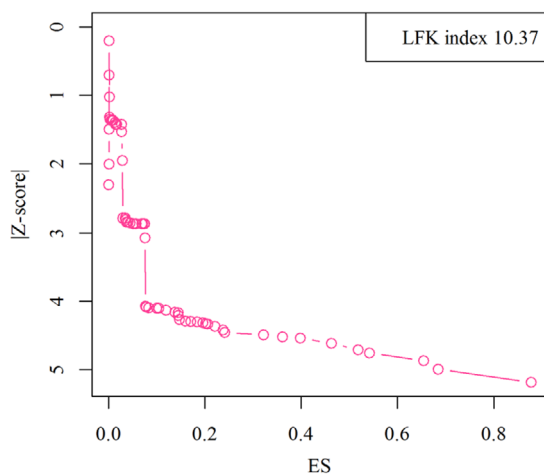


FIGURE 7 Doi plot of the global prevalence for *Trichinella* in pigs. A Luis Furuya-Kanamori (LFK) index 10.37 indicates major asymmetry.

has relatively recently been classified as a lower middle-income country. However, it must be noted that the published date of the included studies goes back to nearly 21 years and the fluctuations in HDI and country income classification level must be considered.

The subgroup analysis also suggested that regions with a tropical wet climate, higher humidity, higher average temperature, and higher annual rainfall had higher prevalence, while the regression indicated that temperature influenced prevalence. These environmental factors could influence *Trichinella* transmission. A previous study suggests that the temperature and humidity, as well as the fauna involved in the decomposition of carcasses, greatly contribute to the longevity of muscle larvae, which have the role of dispersing infection, a function similar to eggs or larvae of other nematodes (Owen & Reid, 2007; Pozio, 2000).

However, we must be careful in interpreting the results regarding environmental parameters, since there are limited studies targeting the survival of muscle larvae in carcasses naturally exposed to different environmental conditions (temperatures, humidity, etc.) (Riva et al., 2012). Also, the findings regarding environment in the meta-analysis presented herein might be influenced by HDI, pig management, and the number of available studies from different regions. For example, Nigeria, with its high prevalence, is located in a climatic region potentially conducive to *Trichinella* transmission; however, it also is a low HDI country with extensive pig rearing systems. In addition, these regional differences are not reflected in the study by Rostami et al. (2018) on global prevalence in wild boar, nor studies on human infection (Devleeschauwer et al., 2015), and studies prior to the 1980s from more temperate climates have reported high prevalence (Kim, 1983).

The management system under which pigs are reared is known to impact parasites and other infections (Delsart et al., 2020; Roepstorff & Nansen, 1994), and the results of the meta-analysis presented herein do not differ from this concept. Indoor, intensive pig management systems can result in lower parasite infections and improve sanitation and food safety (Maes et al., 2020; Roepstorff & Nansen, 1994). In the meta-analysis presented herein, these systems, which predominate in high HDI countries, had lower *Trichinella* prevalence. In contrast, prevalence was higher in pigs kept under a non-intensive system of rearing. In non-intensive management systems (backyard, free roaming or traditional), the housing, feeding supply, and veterinary care can be poor, with pigs scavenging on domestic or agricultural scraps (Nwanta et al., 2011). Given lack of details on some of the production systems in the included studies, the impact of indoor versus outdoor production (separate from intensity) could not be assessed, although studies have shown that outdoor production can increase *Trichinella* exposure (Pozio

et al., 2021). This is an area in which further research is needed, considering the number of outdoor production systems and consumer trends towards free-range or organically produced meat in high HDI countries (Delsart et al., 2020; EFSA Panel on Animal Health and Welfare et al., 2021).

The determination of the pooled prevalence of *Trichinella* in pigs varies depending on the detection method. Higher estimated prevalence was reported in studies using ELISA, alone or in combination with other techniques. Being recommended as a method for surveillance, serological methods could overestimate prevalence due to the persistence of antibodies and false positive cross-reactions (Bruschi et al., 2019; Rostami et al., 2018; Yang et al., 2016). Serological methods such as ELISAs, however, can be more sensitive than direct detection methods such as digestion in animals with mild infection (Nöckler et al., 2005; Yang et al., 2016; Yang et al., 2016;). For individual animal detection and meat inspection, The International Commission on Trichinellosis recommends digestion-based methods (Gajadhar et al., 2019). Digestion methods, though, could underestimate prevalence in low intensity infections. In the studies included in this meta-analysis, approximately half used serological methods and half direct detection methods; however, the majority of the animals were assessed via direct detection methods. Therefore, the global prevalence determined in this meta-analysis is more likely to be an underestimation than an overestimation.

In our findings, *T. spiralis* was found to be the most prevalent *Trichinella* species. However, in many studies, details on the means of confirming the species were limited, and the majority of the studies did not investigate the species present. To better understand the source of *Trichinella* infections in different pig farm settings, especially in outdoor systems, future studies should include species identification. With new molecular methods available and species-specific serological methods, this should be more feasible (Bilska-Zajac et al., 2022; Braasch et al., 2020; Pozio et al., 2020).

The meta-analysis presented herein and the resulting prevalence identified globally and regionally must be understood within the context of its limitations. While there were several studies that reported prevalence based on a combination of diagnostic methods, 26 and 21 relied solely on digestion and ELISA, respectively. Digestion can underestimate prevalence depending on the number of larvae per gram, muscle used, and sample size. Given the standardization of the method, it is the preferred method for meat inspection for human consumption. ELISAs, the recommended method for epidemiological studies, can underestimate prevalence in early stages of infection and overestimate prevalence in later infections with antibodies remaining after the larvae are no longer viable. Combining ELISA with other methods is preferred to obtain a definitive presence of *Trichinella*. An analysis of the data based on sensitivity and specificity of the method could have been beneficial in understanding true prevalence. However, many studies did not report these data, although most of the ELISA-based studies used kits such as ID Screen® *Trichinella* Indirect Multi-species (ID Vet, France) and PrioCheck *Trichinella* Ab (Prionics, Switzerland) which have published sensitivities and specificities and rely on excretory/secretory antigens.

Combining studies using these different diagnostic methods to determine global prevalence could result in some under- and over-estimation. However, the trend was towards the digestion method with the most number of studies and the most number of tested pigs (716,292,004 pigs with digestion vs. 14,579 with ELISA only). In case of regional and country analysis, the impact of diagnostic method likely had more influence on the global prevalence than other variables. For example, all studies from Nigeria, which had the highest prevalence, relied on ELISAs with no supplemental testing. This likely influenced the finding of higher prevalence in Africa. The impact of data from a single country on regional prevalence was potentially exacerbated by the overall lack of current studies or few studies from specific regions. Despite these limitations, our study provides the most comprehensive estimates of the prevalence of *Trichinella* in domestic pigs from a global perspective and highlights the need for more regional studies.

In conclusion, the results of this systematic review and meta-analysis of *Trichinella* in pigs highlight the need of studies in more geographical areas with details on the production system. To effectively prevent and control disease in pig farming, particularly *Trichinella*, adequate strategies for animal health and biosecurity measures are necessary and might need to be adapted for intensive outdoor production systems. According to our review, pigs raised in regions with higher temperatures, humidity, and rainfall showed higher prevalence. However, the higher prevalence was likely related to country HDI and the pig rearing system with lower HDI countries and more extensive production systems occurring in these regions. In low HDI countries where biosecurity measures might be more challenging with the production system, focused meat inspection or promoting public awareness on the subject of safe cooking and freezing of fresh pork could potentially decrease the risk of human infection. A comprehensive One Health approach is highly recommended regarding surveillance and control of *Trichinella* parasites as food-borne zoonoses.

AUTHOR CONTRIBUTIONS

Jennifer K. Ketzis, Milad Badri, Luis Manuel Madeira de Carvalho, and Aida Vafae Esahi designed the study. Amir KarimiPourSaryazdi and Masoud Foroutan searched for primary publications, screened, and appraised primary studies. Amir KarimiPourSaryazdi extracted the data. Jennifer K. Ketzis, Luis Manuel Madeira de Carvalho, Md Robiul Karim, Milad Badri, and Aida Vafae Esahi wrote and edited the manuscript. Meysam Olfatifar contributed to data analysis. All authors read the manuscript and participated in the preparation of the final version of the manuscript.

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CONFLICT OF INTEREST

The authors declare no conflict of interests.

DATA AVAILABILITY STATEMENT

Files containing the data extracted from included manuscripts are available from the corresponding author on request.

ETHICS STATEMENT

This study was approved by the Ethics Committee for Research at Qazvin University of Medical Sciences. The authors confirm that the ethical policies of the journal, as noted on the journal's author guidelines page, have been adhered to and the appropriate ethical review committee approval has been received. While ethical approval was obtained from the university, no animals were used in this study.

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