Global prevalence and factors affecting the level of *Cryptosporidium* contamination in soil: A systematic review, meta-analysis, and meta-regression

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ABSTRACT

Soil contamination with Cryptosporidium is a serious environmental and public health concern. In this systematic review and meta-analysis we estimated the global prevalence of Cryptosporidium contamination in soil and evaluated its association with climatic and hydrometeorological factors. PubMed, Web of Science, Science Direct, China National Knowledge Infrastructure, and Wanfang were searched from database inception up to 24 August 2022. The initial search identified 3220 studies, of which 14 met the inclusion criteria. The results were pooled using a random effects model, and the statistical heterogeneity among the included studies was examined using Cochrane's Q test and I2 statistic. The estimated pooled global prevalence of Cryptosporidium in soil across all studies was 8.13 % (95 % confidence interval, 1.54–18.44). Meta-regression and subgroup analyses showed that Cryptosporidium prevalence in soil was significantly influenced by continent ($p = 0.0002$; $R2 = 49.99$ %), air pressure ($p = 0.0154$; $R2 = 24.01$ %), temperature ($p = 0.0437$; $R2 = 14.53$ %), and detection method ($p = 0.0131$; $R2 = 26.94$ %). These results highlight the need for increased surveillance of Cryptosporidium in soil and its risk factors to inform future development of environmental control interventions and public health policies.

1. Introduction

The zoonotic protozoan parasite Cryptosporidium is a major source of environmental contamination and a threat to human and animal health (Current and Garcia, 1991). Cryptosporidium was first reported to infect humans in the 1970s, however it is currently considered a global food- and water-borne parasite that can cause diarrhea and other extra-intestinal manifestations in humans (Sing et al., 2003; Tang et al., 2021). In 2017, the GBD Diarrhoeal Diseases Collaborators reported that Cryptosporidium is one of the four leading causes of death in children under five (Troeger et al., 2017).

Cryptosporidium outbreaks occur in developed and developing countries (Costa et al., 2022; Ethelberg et al., 2005; Hoxie et al., 1997; Insulander et al., 2008).

Cryptosporidium oocysts are immediately infectious because they do not need to sporulate outside the host (Power et al., 2005). This feature together with the high infectivity of Cryptosporidium, as few as 1-10 oocysts can infect humans (O'Donoghue, 1995), facilitates spreading of infection to new hosts. Cryptosporidium can be transmitted through contaminated water, food, soil, or by direct contact with infected hosts (Fayer, 2004; Ryan et al., 2018). When hosts ingest infectious Cryptosporidium oocysts, sporozoites are released and invade the intestinal epithelium and multiply (Smith et al., 2006). This can lead to diarrhea and dehydration, which can be fatal if left untreated (Stensvold et al., 2015). Individuals who are immunocompromised, such as those living with acquired immunodeficiency syndrome are more susceptible to Cryptosporidium infection (Inungu et al., 2000).

Although Cryptosporidium is largely a water-borne parasite, soil has garnered increased attention as a source and vehicle for Cryptosporidium contamination in different regions of the world (Barwick et al., 2003; Ferreira et al., 2017; Hong et al., 2014; Mandarino-Pereira et al., 2010; Rimhanen-Finne et al., 2001). In light of the growing interest in understanding the epidemiology of Cryptosporidium in soil, we undertook a systematic review to estimate the global prevalence of Cryptosporidium in soil and evaluate the factors that may influence the level of soil contamination with Cryptosporidium, including geographic region (continent), air pressure, temperature, detection method, solar insolation, wind speed, precipitation, humidity, and year of publication.

2. Materials and methods

2.1. Literature search strategy

We searched articles published in five online bibliographic databases from inception up to August 24, 2022, including Web of Science, PubMed, Science Direct, China National Knowledge Infrastructure (CNKI), and Wanfang Data. We used a combination of medical subject headings (MeSH) terms and keywords (e.g., "Cryptosporidium,""soil") in the search strategy (Table S1). The search was not restricted by the publication year and geographic origin of the articles. However, the search was limited to literature published in Chinese or English.

2.2. Article selection and data extraction

The inclusion criteria included original research articles and crosssectional studies about the prevalence of Cryptosporidium contamination in soil and contain information about the number of positive and total samples. Reviews, conference abstracts, books, or articles that had no relevance to the topic were excluded. Also, articles with a sample size of <10 were excluded.

Two authors (M-R. Z. and X-T. L.) conducted an initial search of the databases, screened the titles and abstracts, and downloaded potentially eligible articles. J-P.W. and R-Z. X. independently assessed the full texts of potentially eligible articles based on the inclusion criteria. Divergences were resolved by a third investigator (W. C) and any further disagreements were resolved by consulting H.E. Every study was reviewed based on the publication year, method used for detection of Cryptosporidium in soil, geographic information (continent), climate factors (air pressure, temperature, solar insolation, wind speed, precipitation, humidity). Information on climatic factors was obtained from the National Aeronautics and Space Administration (NASA) Power Data Access Viewer. All information was recorded using Microsoft Excel 2019 (Microsoft, Redmond, WA, USA).

2.3. Assessment of the study quality

The quality of included studies was assessed by one author (M-R. Z.) using the Grading of Recommendations Assessment, Development, and Evaluation (GRADE) approach (Guyatt et al., 2008). The quality assessment included five criteria: defined sample size, publication year, climatic factors (air pressure, temperature, solar insolation, wind speed, precipitation, humidity), a clear description of the detection method, and geographic region of the study, with each criterion represented by one point. The maximum score for all items was 5 points, and studies that met all five criteria received a score of 5. Studies were rated as low (0–1 points), moderate (2–3 points), or high quality (4–5 points) based on their score (Gong et al., 2020).

2.4. Statistical analysis

All relevant data were analyzed using R software version 4.1.3 and calculations were performed using the "meta" R package. Before conducting the meta-analysis, the data were transformed into Gaussian distributions using four transformation methods: log-normal (PLN), logit (PLOGIT), arcsine (PAS), and double arcsine (PFT). Data normality was tested using the Shapiro-Wilk test, with a value of W close to 1 (the maximum value of this statistic) and a p-value ≥0.05 indicating a closer approximation to a Gaussian normal distribution.

A random-effects model based meta-analysis was performed to calculate the pooled prevalence percentage and examine the effect of a single study on the overall pooled prevalence estimate. The magnitude of the between-study heterogeneity was examined using Cochrane's Q test or I^2 statistic. A percentage of I^2 statistic >75 % and $p < 0.10$ suggest significant heterogeneity (Deeks et al., 2019). The results of the meta-analysis were visualized using forest plots. Publication bias was examined by visual inspection of inverted funnel plot asymmetry. Additionally, Egger's test was performed to assess the small study effects. $p < 0.05$ was set at the statistical significance threshold.

Meta-regression and subgroup (covariate-effect) analyses were performed to identify the differences in prevalence between groups and to investigate potential sources of heterogeneity. The covariates were stratified into subgroups by continent (Asia vs Africa

vs North America vs South America vs Europe vs Antarctica); air pressure (kPa) ($90 < x \leq 95$ vs
$95 < x \leq 100$ vs $100 < x \leq 105$); temperature (°C) ($-10 < x \leq 0$ vs $0 < x \leq 10$ vs $10 < x \leq 20$ vs $20 < x \leq 30$); detection method (serologic-based vs nucleic acid-based vs phenotypic-based); solar insolation
(kW-hr/m^2/day) ($2 < x \leq 3$ vs $3 < x \leq 4$ vs $4 < x \leq 5$); wind speed (m/ s) ($0 < x \leq 2$ vs $2 < x \leq 4$ vs $4 < x \leq 6$); precipitation (mm/day) ($0 < x \leq 2$ vs $2 < x \leq 4$ vs $4 < x \leq 6$); humidity (g/kg) ($0 < x \leq 5$ vs
$5 < x \leq 10$ vs $10 < x \leq 15$ vs $15 < x \leq 20$); and publication year divided into two subgroups (from 2000 to 2010 vs after 2010). R packages "ggplot2", "ggmap", "sp", "maps" and "ggsci" were used to display the geographic locations of the included studies on the world's map. The R codes used for data analysis are shown in Table S2.

3. Results

3.1. Characteristics of included studies

The study screening and selection process is summarized in the flow diagram (Fig. 1). A total of 3220 articles were retrieved from five databases. After removing 266 duplicate articles, the titles and abstracts of the remaining articles were reviewed. Of the remaining 2954 articles, 2918 were excluded because they were not relevant to the aim of the review, and the remaining 36 articles were subjected to full-text screening. Of these, 22 were excluded for the following reasons: 1) the subject of the study was not focused on soil contamination, 2) the prevalence rate could not be obtained and 3) the language was not English or Chinese. Finally, 14 studies that met all inclusion criteria and the requirements for systematic review and meta-analysis were included in the analysis. Of the 14 articles included, 11 were high-quality literature and 3 were medium-quality literature based on quality standards. The main characteristics of the included studies are summarized in Table 1.

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3.2.

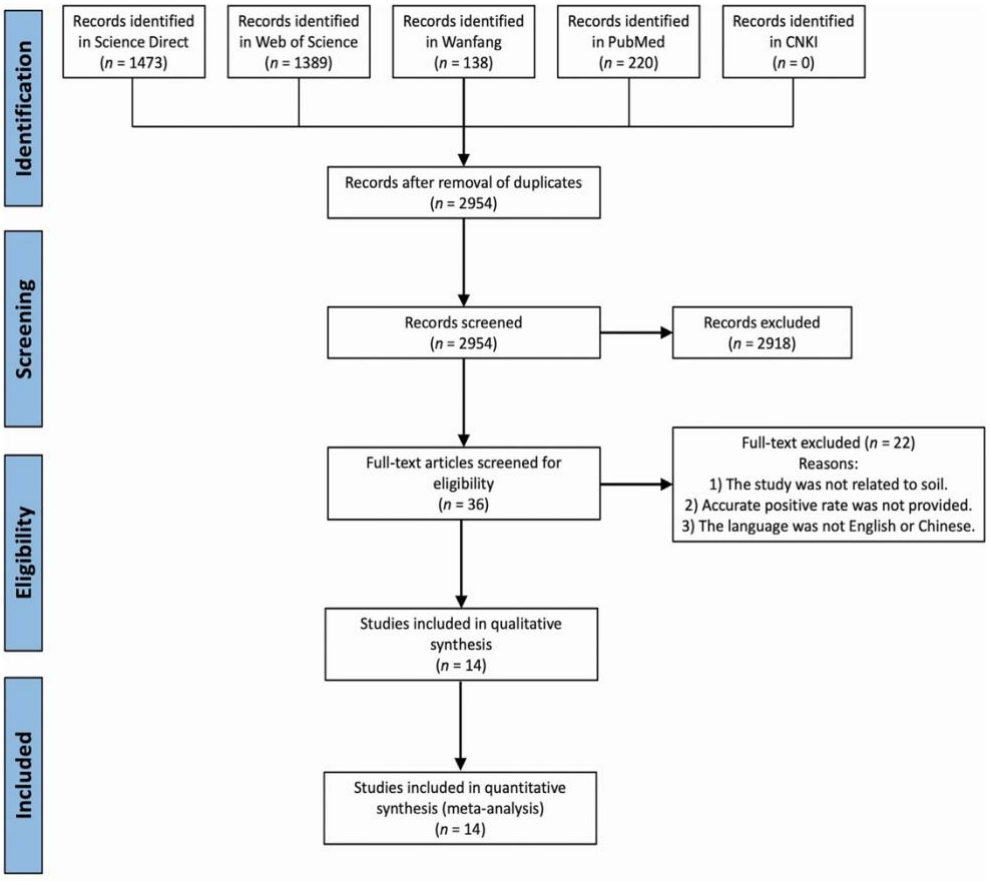

Fig. 1. Preferred Reporting Items for Systematic reviews and Meta-Analysis (PRISMA) flow diagram showing study selection.

Publication heterogeneity, bias and sensitivity analysis

The use of the random effects model was appropriate for the metaanalysis given the substantial heterogeneity between the studies (I^2 = 98 %; τ^2 = 0.0758; $p < 0.01$). The funnel plot showed that all studies are distributed symmetrically, indicating absence of publication bias (Fig. 2). The results of Egger's test further confirmed the lack of statistically significant evidence for publication bias between included studies (p = 0.9828; Fig. 3). The sensitivity analysis did not reveal any significant effects on the overall pooled estimate, after excluding one study at a time, indicating the reliability of the results (Fig. S1).

3.3. Pooled global prevalence of Cryptosporidium in soil

The highest prevalence was detected in the USA (54 %), followed by Mexico (52.3 %), while the lowest prevalence was detected in Antarctica (1.1 %). Interestingly, the prevalence rate was 0 % in some countries, including Brazil, Portugal, Romania, and

Vietnam. The overall pooled prevalence of Cryptosporidium oocysts in soil was estimated to be 8.13 % (95 % confidence interval (CI), 1.54–18.44) (Fig. 4).

3.4. Meta-regression analysis of study-level covariates

Meta-regression and subgroup analyses were used to investigate whether statistical heterogeneity between the pooled prevalence estimate is related to study-level covariates (continent, air pressure, temperature, detection method, solar insolation, wind speed, precipitation, humidity, and publication year). The results of meta-regression analysis showed statistically significant relationship between the pooled prevalence estimate and study-level covariates continent, air pressure, temperature, and detection method.

3.4.1. Prevalence of Cryptosporidium by continent

As shown in Table 2, the highest prevalence of Cryptosporidium in soil samples was found in North America 39.18 % (95 % CI, 15.50–65.85; I^2 = 99 %; τ^2 = 0.0513; $p < 0.01$); followed by Asia 11.24 % (95 % CI, 0.00–57.30; I^2 = 93 %; τ^2 = 0.1170; $p < 0.01$); Europe 2.89 % (95 % CI, 0.20–7.55; I^2 = 56 %; τ^2 = 0.0053; p = 0.08); and South America 0.00 % (95 % CI, 0.00–0.0011; I^2 = 0 %; τ^2 = 00; p = 0.77). Fig. 5 shows a geographic information system (GIS) map displaying the geographic locations where Cryptosporidium was detected in soil samples worldwide. The forest plot comparing prevalence among continents is shown in Fig. S2. Meta-regression analysis showed that prevalence of Cryptosporidium in soil was significantly affected by North America and South America (p = 0.0002; R^2 = 49.99 %).

3.4.2. Prevalence of Cryptosporidium by air pressure

The prevalences of Cryptosporidium in the soil at different air pressure ranging from 90 kPa < x ≤ 95 kPa vs 95 kPa < x ≤ 100 kPa vs 100 kPa < x ≤ 105 kPa are shown (Table 2; Fig. S3). The highest estimated prevalence of Cryptosporidium, 55.12 % (95 % CI, 47.77–62.36; I^2 = 16 %; τ^2 = 0.0008; p = 0.31), was detected at 90 kPa < x ≤ 95 kPa, whereas the lowest estimated prevalence of Cryptosporidium, 10.80 % (95 % CI, 0.00–32.41; I^2 = 88 %; τ^2 = 0.1022; $p < 0.01$), was detected at 100 kPa < x ≤ 105 kPa. This result suggests that prevalence of Cryptosporidium in soil was significantly affected by air pressure at 90 kPa < x ≤ 95 kPa (p = 0.0154; R^2 = 24.01 %).

3.4.3. Prevalence of Cryptosporidium based on temperature

prevalence of Cryptosporidium in the soil at temperatures ranging from −10 °C < x ≤ 0 °C vs 0 °C < x ≤ 10 °C vs 10 °C < x ≤ 20 °C vs 20 °C < x ≤ 30 °C is shown (Table 2; Fig. S4). The estimated prevalence of Cryptosporidium at −10 °C < x ≤ 0 °C was the highest 32.65 % (95 % CI, 7.74–62.51; I^2 = 71 %; τ^2 = 0.0811; $p < 0.01$); and at 20 °C < x ≤ 30 °C was the lowest 6.58 % (95 % CI: 0.00–27.75; I^2 = 98 %; τ^2 = 0.1238; $p < 0.01$). These results indicate that prevalence of Cryptosporidium in soil was

significantly affected by temperature when the temperature reached 20 °C < x ≤ 30 °C ($p = 0.0437$; $R^2 =$ 14.53 %).

Table 1 Summary of the main characteristics of the studies included in the meta-analysis.

Reference	Score	No. positive/total examined (%)	Detection method(s)	Country
Rimhanen-Finne et al., 2001	5	3/44 (6.8 %)	Nucleic acid based	Finland
Barwick et al., 2003	3	133/782 (17 %)	Phenotypic-based	United States
Boyer and Kuczynska, 2010	5	175/324 (54 %)	Serologic-based	United States
Mandarino-Pereira et al., 2010	5	0/125 (0 %)	Phenotypic-based	Brazil
Dado et al., 2012	3	38/625 (6 %)	Nucleic acid based	Spain
Balderrama-Carmona et al., 2014	5	11/21 (52.3 %)	Serologic-based	Mexico
Hong et al., 2014	5	11/34 (32.3 %)	Nucleic acid-based	Korea
Tudor, 2015	5	0/45 (0 %)	Phenotypic-based	Romania
Ferreira et al., 2017	5	0/18 (0 %)	Phenotypic-based	Portugal
da Silva et al., 2016	5	0/600 (0 %)	Serologic-based	Brazil
Lim et al., 2018	3	2/175 (1.1 %)	Phenotypic-based	Antarctica
Capone et al., 2020	5	23/95 (24.2 %)	Nucleic acid-based	Mozambique
Lee et al., 2021	5	0/83 (0 %)	Phenotypic-based	Brazil
Tram et al., 2022	5	0/21 (0 %)	Phenotypic-based	Vietnam

3.4.4. Prevalence of Cryptosporidium based on detection method

The methods used for detection of Cryptosporidium are generally grouped into three broad categories: nucleic acid-based, phenotypicbased, and serologic-based methods. The estimated prevalence of Cryptosporidium based on nucleic acid-based detection methods was 15.20 % (95 % CI, 4.82–29.62; $I^2 = 92$ %; $\tau^2 = 0.0267$; $p < 0.01$); phenotypicbased methods was 1.11 % (95 % CI, 0.00–5.59; $I^2 = 96$ %; $\tau^2 = 0.0196$; $p < 0.01$); and serological methods was 26.64 % (95 % CI, 0.00–77.59; $I^2 = 100$; $\tau^2 = 0.2114$; $p < 0.01$). The prevalence of Cryptosporidium in soil by detection method is shown (Table 2; Fig. S5). The meta-regression analysis showed that prevalence of Cryptosporidium in soil was significantly influenced by phenotypic-based detection methods ($p = 0.0131$; $R^2 =$

26.94 %).

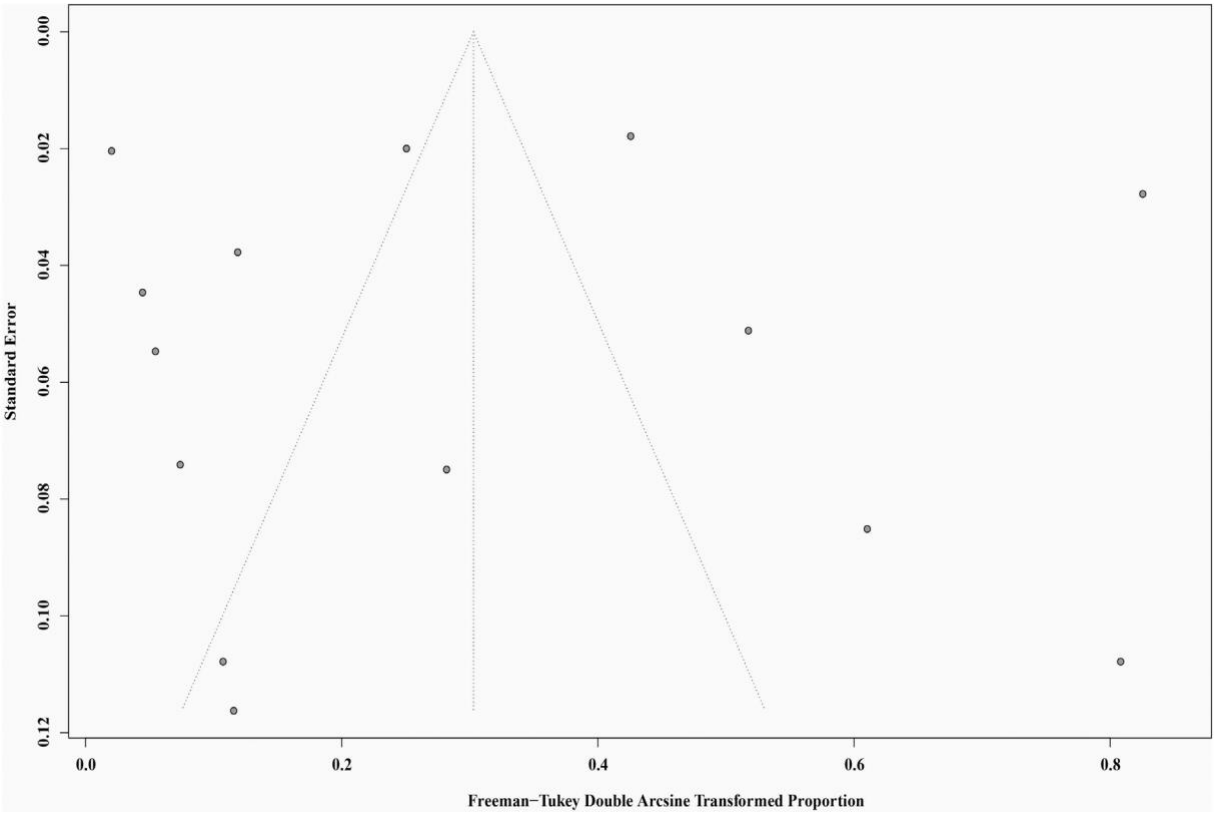

Fig.2. Funnel plot of the Freeman-Tukey double arcsine transformed prevalence of Cryptosporidium in soil examining publication bias. Each dot represents a different study.

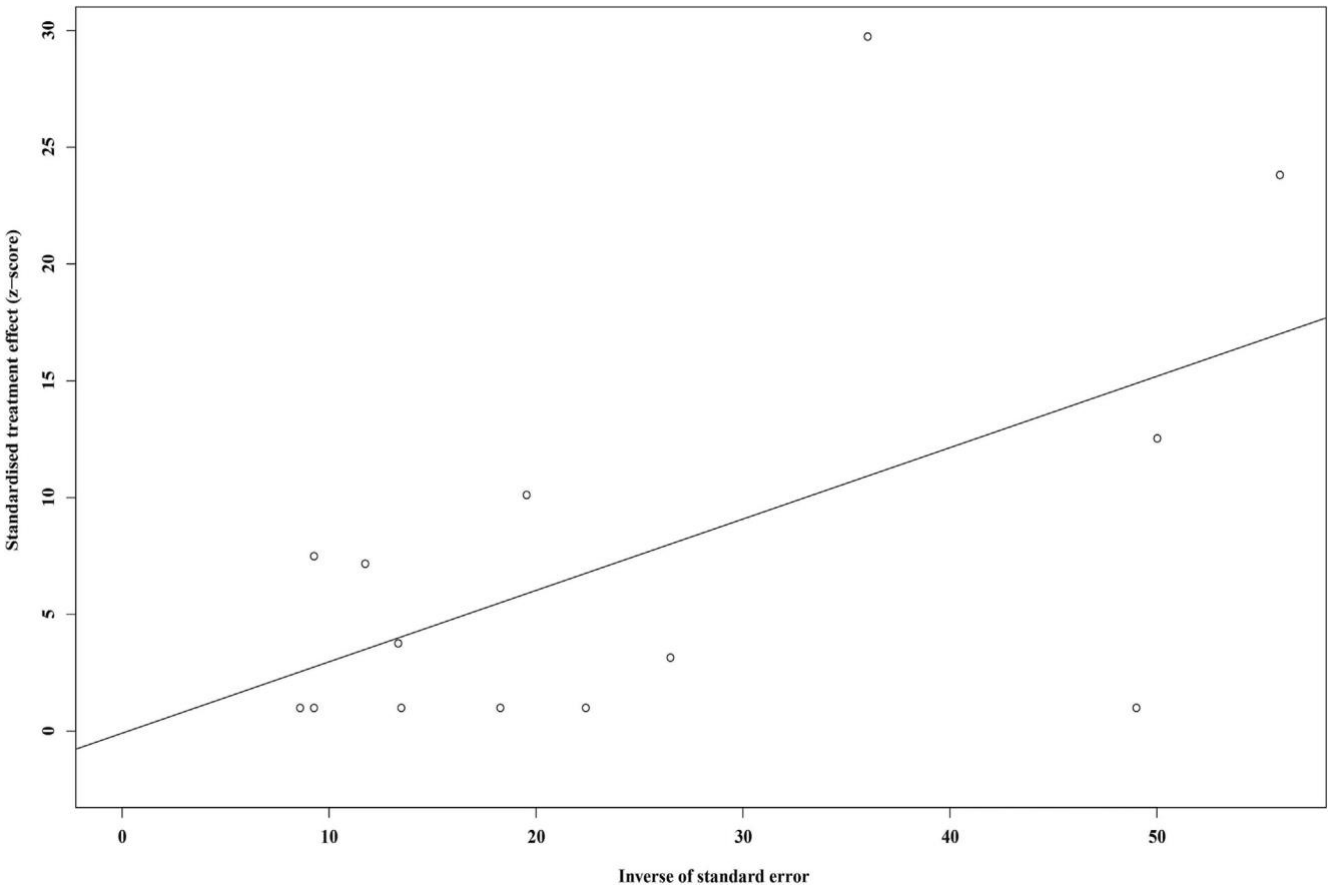

Fig. 3. Egger's plot examining the publication bias between the included studies.

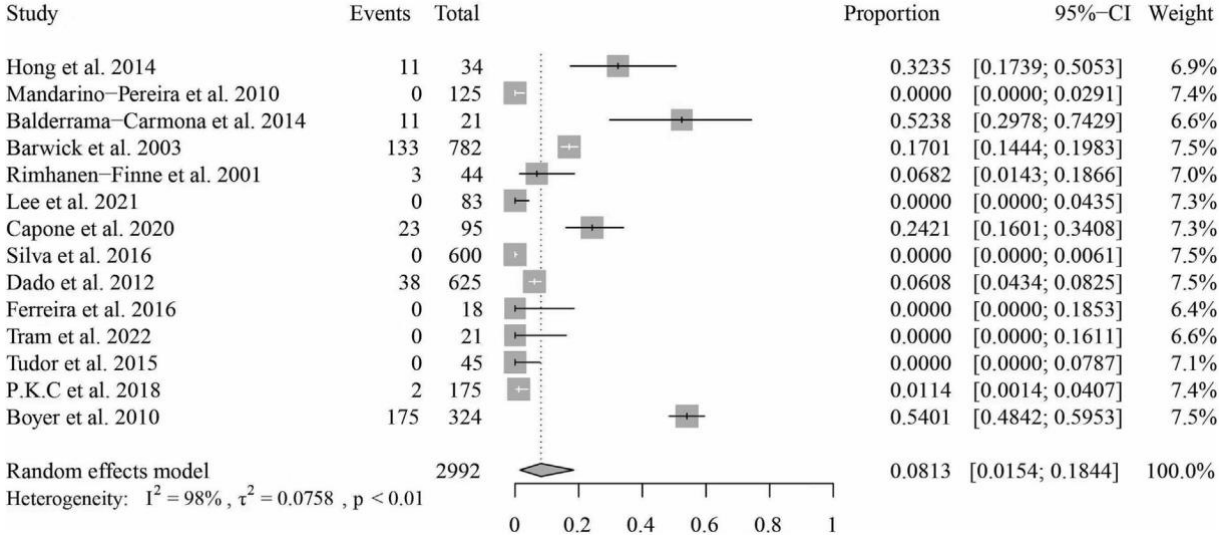

Fig. 4. Forest plot of pooled prevalence of Cryptosporidium in soil. Gray squares and the horizontal lines are the point estimates and 95 % confidence interval (CI). The diamond represents the pooled estimate (width denotes 95 % CI).

3.4.5. Prevalence of Cryptosporidium based on solar insolation

The prevalence of Cryptosporidium in the soil at different ranges of solar insolation values: 2 kW-hr/m^2/day < x ≤ 3 kW-hr/m^2/day vs 3 kW-hr/m^2/ day < x ≤ 4 kW-

hr/m^2/day vs 4 kW-hr/m^2/day < x ≤ 5 kW-hr/m^2/day vs 5 kW-hr/m^2/day < x ≤ 6 kW-hr/m^2/day is shown (Table 2; Fig. S6). The estimated prevalence of Cryptosporidium at 2 kW-hr/m^2/day < x ≤ 3 kWhr/m^2/day was the highest 20.67 % (95 % CI: 2.85–45.37; I^2 = 89 %; τ^2 = 0.1077; p < 0.01), whereas at 5 kW-hr/m^2/day < x ≤ 6 kW-hr/m^2/ day was the lowest 16.40 % (95 % CI, 0.00–58.17; I^2 = 99 %; τ2 = 0.2021; p < 0.01). This result suggests that prevalence of Cryptosporidium in soil was not significantly affected by solar insolation (p = 0.6216 > 0.05; R^2 = 0.00 %).

3.4.6. Prevalence of Cryptosporidium based on wind speed

The prevalence of Cryptosporidium in the soil at different wind speed values ranging from 0 m/s < x ≤ 2 m/s vs 2 m/s < x ≤ 4 m/s vs 4 m/s < x ≤ 6 m/s are obtained (Table 2; Fig. S7). The estimated prevalence of Cryptosporidium was highest 32.19 % (95 % CI, 13.22–54.72; I^2 = 99 %; τ^2 = 0.1270; p< 0.01) at 0 m/s < x ≤ 2 m/s, whereas the lowest prevalence 5.22 % (95 % CI, 0.00–24.65; I^2 = 93 %; τ^2 = 0.0547; p < 0.01) was detected at 4 m/s < x ≤ 6 m/s. This result suggests that prevalence of Cryptosporidium in soil was not significantly affected by wind speed (p = 0.2022; R^2 = 3.15 %).

Table 2 Pooled prevalence, subgroup analysis and heterogeneity statistics of Cryptosporidium in soil.

Variab		% (95 % CI)	Heterogeneity statistics		Univariate meta-		
			p–value	I2	p–value	Coefficient (95 % CI)	R^2 (%)
Continent	Asia	11.24 (0.00–57.30)	<0.01	93 %	0.0002	0.4712 (0.2214–0.7210)	49.99
	Europe	2.89 (0.20–7.55)	0.08	56 %			
	South America	0.07 (0.00–1.73)	0.77	0 %			
	North America	39.18 (15.50–65.85)	<0.01	99 %			
Air pressure (kPa)	90 < x ≤ 95	55.12 (47.77–62.36)	0.31	16 %	0.0154	0.4248 (0.0813–0.7683)	24.01
	95 < x ≤ 100	14.89 (0.49–49.88)	<0.01	98 %			
	100 < x ≤ 105	10.80 (0.00–32.41)	<0.01	88 %			

Temperature (°C)	−10 °C < x ≤ 0 °C	32.65 (7.74–62.51)	<0.01	71 %	0.0437	−0.3258 (−0.6424–−0.0092)	14.53
	0 °C < x ≤ 10 °C	32.13 (6.14–66.08)	<0.01	94 %			
	10 °C < x ≤ 20 °C	25.57 (1.83–61.62)	<0.01	96 %			
	20 °C < x ≤ 30 °C	6.58 (0.00–27.75)	<0.01	98 %			
Detection method	Nucleic acid-based	15.20 (4.82–29.62)	<0.01	92 %	0.0131	−0.3037 (−0.5438–−0.0637)	26.94
	Phenotypic-based	1.11 (0.00–5.59)	<0.01	96 %			
	Serologic-based	26.64 (0.00–77.59)	<0.01	100 %			
Solar insolation (kW-hr/m2/day) *	2 < x ≤ 3	20.67 (2.85–45.37)	<0.01	89 %	0.6216	−0.1001 (−0.4977–0.2974)	0.00
	4 < x ≤ 5	19.51 (1.52–48.90)	<0.01	97 %			
	5 < x ≤ 6	16.40 (0.00–58.17)	<0.01	99 %			
Wind Speed (m/s)	0 < x ≤ 2	32.19 (13.22–54.72)	<0.01	99 %	0.2022	−0.2577 (−0.6537–0.1383)	3.15
	2 < x ≤ 4	13.39 (0.00–46.23)	<0.01	83 %			
	4 < x ≤ 6	5.22 (0.00–24.65)	<0.01	93 %			
Precipitation (mm/day)	0 < x ≤ 2	23.90 (2.12–54.34)	<0.01	90 %	0.1830	0.4761 (−0.2247–1.1770)	4.70
	2 < x ≤ 4	18.06 (2.53–42.02)	<0.01	97 %			
	4 < x ≤ 6	12.95 (0.00–45.45)	<0.01	98 %			

Humidity (g/kg)	0 < x ≤ 5	32.65 (7.74–62.51)	<0.01	71 %	0.7968	0.0459 (−0.3034–0.3952)	0.00
	5 < x ≤ 10	19.01 (2.38–44.79)	<0.01	95 %			
	10 < x ≤ 15	25.55 (3.04–58.89)	<0.01	99 %			
	15 < x ≤ 20	0.00 (0.00–1.36)	0.66	0 %			
Publication Year	After 2010	3.45 (0.00–14.08)	<0.01	95 %	0.4918	−0.0994 (−0.3828–0.1840)	0.00
	2000–2010	21.35 (0.30–58.70)	<0.01	99 %			

* We could not perform statistical analysis on the range 3 < x ≤ 4 because it encompassed only one study.

Fig. 5. Global map showing the geographic distribution of studies included in the meta-analysis.

3.4.7. Prevalence of Cryptosporidium based on precipitation

We obtained the prevalence of Cryptosporidium in the soil at different precipitation values ranging from 0 mm/day < x ≤ 2 mm/day vs 2 mm/ day < x ≤ 4 mm/day vs 4 mm/day < x ≤ 6 mm/day (Table 2; Fig. S8). The estimated prevalence of Cryptosporidium at 0 mm/day < x ≤ 2 mm/ day was the highest 23.90 % (95 % CI, 2.12–54.34; I^2 = 90 %; τ^2 = 0.1176; p < 0.01), whereas at 4 mm/day < x ≤ 6 mm/day was the lowest 12.95 % (95 % CI, 0.00–45.45; I^2 = 98 %; τ2 = 0.1308; p < 0.01). This result suggests that prevalence of Cryptosporidium in soil was not significantly affected by precipitation (p = 0.1830; R^2 = 4.70 %).

3.4.8. Prevalence of Cryptosporidium based on humidity

The prevalence of Cryptosporidium in the soil at different humidity ranges, including 0 g/kg < x ≤ 5 g/kg vs 5 g/kg < x ≤ 10 g/kg vs10 g/kg < x ≤ 15 g/kg vs 15 g/kg < x ≤ 20 g/kg are shown (Table 2; Fig. S9). The estimated prevalence of Cryptosporidium at 0 g/kg < x ≤ 5 g/kg was the highest 32.65 % (95 % CI, 7.74–62.51; I^2 = 71 %; τ^2 = 0.0811; p < 0.01), whereas at 15 g/kg < x ≤ 20 g/kg was the lowest 0.00 % (95 % CI, 0.00–1.36; I^2 = 0 %; τ^2 = 0; p = 0.06). This result suggests that prevalence of Cryptosporidium in soil was not significantly affected by humidity (p = 0.7968; R^2 = 0.00 %).

3.4.9. Prevalence of Cryptosporidium by publication year

Based on the publication year, the prevalence of Cryptosporidium in soil from 2000 to 2010 was 21.35 % (95 % CI, 0.30–58.70; I^2 = 99 %; τ^2 = 0.1487; p < 0.01), compared with 3.45 % (95 % CI, 0.00–14.08; I^2 = 95; τ^2 = 0.0548; p < 0.01) after the year 2010 (Table 2; Fig. S10). This result shows that prevalence of Cryptosporidium in soil was not significantly affected by publication year (p = 0.1540; R^2 = 9.26 %).

4. Discussion

Cryptosporidium is a leading cause of parasitic diarrhea particularly in immunocompromised individuals (Widmer et al., 2020) and is prevalent in developing countries (Areeshi et al., 2007; Yang et al., 2021). In addition to the lack of vaccine, available treatments for cryptosporidiosis are not effective (Sparks et al., 2015). Accurate estimates of the prevalence of Cryptosporidium in the environment are therefore needed to inform infection control policies, allocate resources, and guide further research to improve knowledge of the epidemiology of Cryptosporidium. In this review, the estimated pooled global prevalence of Cryptosporidium in soil was 8.13 % (95 % CI, 1.54–18.44). There were significant heterogeneities between studies (I^2 =

98; τ^2 = 0.0758; $p < 0.01$), highlighting the need to identify the factors which can influence the prevalence of Cryptosporidium in soil.

Heterogeneities between studies could be partially explained by metaregression analysis, which revealed significant differences in the prevalence of Cryptosporidium in soil between studies from different continents. Cryptosporidium seroprevalence increases with economic deprivation, social inequality and decreases with educational attainment (Becker et al., 2015), suggesting that Cryptosporidium disparities between continents may be influenced by social determinants of health. Low socioeconomic status is often associated with lack or insufficient resources needed for proper sanitation or educational avoidance of transmission routes through exposure to infected animals or contaminated food and water. Therefore, better understanding of the interactions between socioeconomic and environmental factors influencing the epidemiology of Cryptosporidium particularly in low- and middle-income countries is necessary to guide Cryptosporidium infection control interventions in resource-limited regions.

The hydrometeorological variable air pressure had a significant effect on the prevalence of Cryptosporidium in soil ($p = 0.0154$; R^2 = 24.01 %). Air pressure is generally considered a proxy for storm activity. For example, with low pressure, commonly associated with clouds, rain and wind, soil moisture increases by rainfall, which improves the survival of Cryptosporidium outside the host (Colston et al., 2022). Temperature explained part of the heterogeneity between the included studies and prevalence of Cryptosporidium in soil was negatively associated with temperature. While oocysts survival increases at temperatures <15 °C in soil (Jenkins et al., 2002), exposure to higher temperatures (>25 °C) and increased UV-A/B insolation can cause oocyst degradation (King et al., 2008; Olson et al., 1999; Robertson et al., 1992). The results of the present and previous studies (Jenkins et al., 2002; King et al., 2008; Olson et al., 1999; Robertson et al., 1992) show that the evidence base for the temporal and spatial response of Cryptosporidium to fluctuations in temperature remains unresolved, partly because of the contrasting results of the impact of temperature on the survival and infectivity of Cryptosporidium oocysts in the environment (Ikiroma and Pollock, 2021; Peng et al., 2008; Wang et al., 2023).

The detection methods explained some of the heterogeneity among studies included in this review ($p = 0.0131$; R^2 = 26.94 %). The presence/absence of Cryptosporidium is generally achieved by using microscopy (phenotypic-based), serological assays, or molecular methods (nucleic acid-based). Using various methodologies makes a direct comparison between studies and geographical regions difficult. The estimated prevalence was highest based on the serologic-based detection methods (26.64 %), followed by the nucleic acid-based detection methods (15.20 %), while phenotypic-based detection methods had the lowest prevalence (1.11 %). Prevalence based on microscopy is lower than that obtained by molecular methods (Mwingira et al., 2014). The identification of Cryptosporidium species by light microscopy is unreliable and not specific, as most Cryptosporidium spp. have similar morphological attributes (Fall et al., 2003). Serologic assays are less labor intensive and provide better sensitivity compared

with microscopic examination (Josko, 2012). Nucleic acid-based detection of Cryptosporidium offers more advantages, including better sensitivity and specificity, and identification of Cryptosporidium to the specie/genotype/ sub-genotype levels, which can be invaluable for the understanding of the parasite epidemiology and for investigation of outbreaks (BonninJusserand et al., 2019; Coetzee et al., 2008; Hassan et al., 2021; McLauchlin et al., 2000; Xiao et al., 2004).

In the present study, solar insolation, wind speed, precipitation, humidity, and publication year had no significant influence on the prevalence of Cryptosporidium in soil. The effects of these and other climatic variables on the survival, infectivity, and distribution of Cryptosporidium oocysts in the environment have been extensively studied but is a multifactorial issue that may not be easily resolved (Colston et al., 2022; Ikiroma and Pollock, 2021; Wang et al., 2023). Although no significant association was detected between precipitation and Cryptosporidium prevalence in our review and despite the widely divergent evidence in the literature (Ikiroma and Pollock, 2021; Wang et al., 2023), the incidence of Cryptosporidium increases significantly after flooding (Bunyavanich et al., 2003) and rainfall can facilitate the dispersal of Cryptosporidium oocysts in soil, increasing the risk for transmission of infection (Ramirez et al., 2009). Therefore, the development of methods with good sensitivity and specificity for early detection of Cryptosporidium is soil and identification of contaminated food or water can reduce the risk of human infection.

We employed stringent inclusion criteria and sound approach consistent with the PRISMA statement to search five major electronic bibliographic databases. However, our study has some limitations. First, literature search did not include all databases (e.g., Cochrane Library, Embase, Google Scholar, Scopus). Second, the number of studies that met the inclusion criteria are too small to allow an accurate synthesis of the global prevalence of Cryptosporidium in soil. Third, our search did not include the gray literature (e.g., reports from government agencies, academic institutions, or private organizations) and information was not available for all regions of the world, which limited the analysis we were able to perform. Fourth, certain factors such as soil characteristics and climatic parameters were not fully reported in all studies. Fifth, only studies published in English or Chinese language were included, which further limited the number of studies included in the meta-analysis.

5. Conclusion

This systematic review has estimated 8.13 % global pooled prevalence of Cryptosporidium contamination in soil and revealed significant association of continent, air pressure, temperature, and method of detection with Cryptosporidium prevalence. Considering that Cryptosporidium outbreaks occur in many parts of the world, the development of more efficient methods for early and accurate detection of Cryptosporidium in soil is critical for implementation of infection control measures to prevent transmission. Although having better detection methods is an essential part of this jigsaw, any Cryptosporidium control strategies should also consider implementation

of hygienic measures such as drinking safe water and washing vegetables and fruits. Our findings highlight gaps in existing evidence regarding the performance/accuracy of Cryptosporidium detection methods and the effects of fluctuations of environmental/climatic/hydrometeorological factors on the epidemiology of Cryptosporidium in soil, providing new insight to guide future research studies. Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2023.164286.

CRediT authorship contribution statement

Mu-Ran Zuo: Formal analysis, Investigation, Methodology, Writing – original draft. Xiao-Ting Li: Formal analysis, Investigation, Methodology. Rui-Zhe Xu: Formal analysis, Investigation, Methodology. Wen-Chao Sun: Conceptualization, Writing – original draft. HanyM.Elsheikha: Conceptualization, Validation, Resources, Writing – review & editing. Wei Cong: Conceptualization, Investigation, Methodology, Validation, Supervision, Project administration, Resources, Writing – review & editing.

Data availability

No data was used for the research described in the article

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this article.

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