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Disentangling the transmission of *Clonorchis sinensis*: A one health approach

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ABSTRACT

Background: Clonorchiasis, a zoonotic parasitic disease caused by *Clonorchis sinensis*, remains a public health issue in East Asia. The full spectrum of suitable animal hosts has not been clearly delineated, and quantitative assessments systematically linking animal infections to human infection risks remain scarce. This study applied a One Health framework to investigate the role of various domestic animals as reservoir hosts and to quantify their association with human infection risk in an endemic region of China.

Methods: A cross-sectional survey was conducted from October 2024 to March 2025 in five villages in Fusui County, Guangxi Zhuang Autonomous Region, China. We collected fecal samples from 1013 human participants and 802 domestic animals (dogs, cats, cattle, sheep, pigs, chickens, ducks, and geese). A two-level Bayesian hierarchical logistic regression model was employed to assess the association between village-level animal infection status and individual human infection risk, controlling for individual-level demographic and behavioral factors.

Results: The overall prevalence of *C. sinensis* infection in humans was 30.50 % (309/1013, 95 % Confidence Interval (CI): 27.68–33.44 %). Among the eight animal species surveyed, infections were detected only in cats (5/53, 9.43 %; 95 % CI: 3.13–20.66 %) and dogs (1/169, 0.59 %; 95 % CI: 0.01–3.25 %). No infections were found in poultry or livestock. The Bayesian multilevel model revealed that after controlling for individual risk factors, the presence of *C. sinensis* infection in animals within a village was significantly associated with a 2.37-fold increase in the odds of human infection (adjusted odds ratio (aOR) = 2.37, 95 % CI: 1.23–3.58). Key individual-level risk factors included being male (aOR = 3.46, 95 % CI: 2.44–4.98) and having a history of consuming raw freshwater fish (aOR = 5.27, 95 % CI: 3.70–7.48).

Conclusions: Our findings demonstrate that clonorchiasis is an important public health problem in the study area, and cats and dogs are the primary animal reservoir hosts for *C. sinensis* there, while poultry and livestock do not appear to participate in the local transmission cycle. The presence of infected animals in shared communities is significantly associated with an increased risk of human infection.

1. Introduction

Clonorchiasis, caused by *Clonorchis sinensis*, is a food-borne zoonotic parasitic disease endemic in East Asian countries, notably China, South Korea, and Vietnam [1]. Adult worms parasitize in the hepatobiliary system of humans and various mammals, resulting in chronic cholangitis, biliary fibrosis, cholelithiasis, and even cholangiocarcinoma after prolonged infection [2]. Clonorchiasis poses a significant public health concern, especially in China, where recent geospatial modeling estimates indicate approximately 99.13 million individuals at risk and 9.46 million under infection [3].

The life cycle of *C. sinensis* involves multiple hosts. Eggs excreted by definitive hosts (humans and animals such as dogs and cats) are ingested by freshwater snails, developing into cercariae [4–6]. These cercariae subsequently penetrate freshwater fish, encysting in their muscle tissues as metacercariae. Upon ingestion of raw or inadequately cooked freshwater fish by humans, metacercariae excyst in the duodenum, releasing larvae that migrate upwards through the common bile duct and develop

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into hermaphroditic adult worms within the intrahepatic bile ducts [2,7–9]. These adult worms can survive for over 10 years, with documented cases of survival up to 26 years [10]. Each adult worm produces thousands of eggs daily [11], perpetuating the transmission cycle. Although *C. sinensis* is sensitive to antiparasitic drugs such as praziquantel with cure rates reaching up to 98.5 % [12], the persistence of even a small number of definitive or reservoir hosts in the ecosystem enables rapid resurgence of egg contamination once control measures are relaxed, resulting in recurrent epidemics. Humans, along with companion animals such as dogs and cats, are widely recognized as definitive hosts [11,12]. Among livestock species, infections in pigs have been reported [13], however, the suitability of cattle, sheep, and other domestic animals as hosts remains uncertain. Findings on avian hosts are controversial, some studies indicate potential infections in chickens and ducks [14], whereas others failed to detect natural infections in endemic areas [15,16], and experimental infections under laboratory conditions were unsuccessful [16]. Animal hosts, particularly free-roaming domestic cats and dogs within human communities, constitute a major environmental source of *C. sinensis* eggs [13]. The presence of animal hosts complicates efforts for clonorchiasis control [14,15], but the full spectrum of suitable animal hosts has not been clearly delineated due to a lack of systematic epidemiological field data. Furthermore, quantitative assessments systematically linking animal infections to human infection risks remain scarce.

In shared communities, where humans and animals cohabit, sharing water sources, food, and environmental exposure [17], animal hosts potentially exacerbate community-wide exposure through persistent environmental contamination by parasite eggs, thereby influencing human infection risks [18]. Existing epidemiological studies primarily emphasize individual behavioral risk factors [19–21] (e.g., dietary habits and sanitation practices) and less addressed the association between animal and human infections in shared communities. The application of a One Health framework that highlights the dynamic linkages among human, animal, and environmental health can offer critical insights for the design of more effective control strategies targeting clonorchiasis.

This study was conducted in Fusui County, Guangxi Zhuang Autonomous Region, one of the known hyperendemic areas for clonorchiasis [22]. Simultaneous surveys of human and animal infections were performed to systematically evaluate whether animals (dogs, cats, cattle, sheep, pigs, chickens, ducks, and geese) are potential reservoir hosts and their association with human infections within shared communities. Villages were treated as the basic exposure unit, defining the exposure variable as the detection of *C. sinensis* infection in any animal species within the community. Given the hierarchical nature of data (individuals nested within villages), we employed a two-level Bayesian hierarchical logistic regression model. This model quantitatively assessed the impact of village-level animal infections on individual human infection risk, controlling for individual demographic and behavioral confounders. Results of this research are anticipated to inform integrated human-animal-environment “One Health” strategies for clonorchiasis control.

2. Methods

2.1. Study site and design

This study was conducted in Fusui County, Chongzuo City, Guangxi Zhuang Autonomous Region, China. Previous studies have shown Guangxi to be highly endemic for clonorchiasis, where 64 % of counties exhibit infection rates exceeding 10 %. Fusui County reports an exceptionally high prevalence of approximately 40.3 % [22].

A cross-sectional design was adopted, conducted from October 2024 to March 2025, utilizing a “One Health” approach to concurrently assess infections and associated risk factors in both humans and animals. For the human survey, an expected *C. sinensis* prevalence of 15 % and an allowable error of 20 % were assumed, and the sample size increased by 50 % taking consideration of the cluster sampling. This resulted in an overall sampling size of 850, which was adjusted to 1000 finally. A multi-stage cluster sampling design was applied. Fusui County was divided into five geographic parts (east, west, south, north and central), and from each part one township was randomly selected. Then, from each township a village with about 200 villagers was selected at random. If the village had over 200 villagers, a unit of about 200 villagers was separated and surveyed. Eight species of domestic animals (cats, dogs, chickens, ducks, geese, cattle, sheep and pigs) were planned to be included with 20 individuals per species in each village, resulting in an overall size of 800 animals in the county. If fewer than 20 individuals of a given species in one village, the shortfall was supplemented by same species in other village; if fewer than 100 animals of a given species in five villages, the shortfall was supplemented by other species. All animals were selected from the households participating in human survey.

2.2. Sample collection and examination

Approximately 30 g of fecal samples per participant were collected, labeled, and refrigerated at 4 °C for laboratory analysis and information including gender, age, occupation, education, and raw freshwater fish consumption behaviors of the examined individual was also recorded. Fresh animal fecal samples were simultaneously collected, recording species, gender, and age reported by owners. Modified Kato-Katz thick smear techniques were applied to human and animal samples, examining two slides per sample under microscopy [23].

2.3. Questionnaire survey

Structured questionnaires collected demographic data, dietary habits, family information, and animal breeding situation from residents. The questionnaire consisted of two components: a household survey and an individual survey. The household questionnaire assessed family-level practices, including the frequency of preparation, purchasing behaviors, social practices of giving or receiving raw fish, use separate cutting boards for raw and cooked foods, and the keeping of domestic animals such as cats and dogs. To further explore factors associated with *C. sinensis* infection, 40 out of 200 participants in each village were randomly selected for the individual survey, with an overall number of 200 individuals in the county. This component collected the individual's behaviors and frequency of raw freshwater fish consumption, as well as knowledge of clonorchiasis, its transmission routes, and associated health risks.

2.4. Statistical analysis

All analyses were performed using R software (version 4.1.2). Egg counts per gram of feces (EPG) were calculated as the mean egg count per two slides multiplied by 24. EPG data were log-transformed to approximate normality, with geometric means and 95 % confidence intervals (CI) calculated using the Cox method [24]. Descriptive analyses were first conducted to present the prevalence of *C. sinensis* infection among humans (stratified by village, gender, age, occupation, education, and the practice of consuming raw freshwater fish) and domestic animals (stratified by village, gender, and species). Categorical variables were compared via Chi-square or Fisher's exact tests where appropriate.

Logistic regression models were then applied to explore factors associated with *C. sinensis* infection in both humans and domestic animals. For the human analysis, explanatory variables were derived from individual questionnaires and the corresponding household questionnaires. With *C. sinensis* infection as the outcome variable, univariable logistic regression was first performed with candidate predictors including village, gender, age, education, hearing of clonorchiasis, knowing transmission route, knowing harm, have eaten raw freshwater fish, frequency on consuming raw freshwater fish, frequency of making raw freshwater fish, use separate cutting boards for raw and cooked foods, purchased raw freshwater fish from outside, gave raw freshwater fish to neighbors, received raw freshwater fish as a gift, and whether to keep cat/dog/livestock/poultry. Variables with $P < 0.05$ in the univariable analysis were subsequently entered into the multivariable logistic regression model. The analytic procedure for domestic animals was the same as that for humans.

Subsequently, to assess the impact of animal infections on human infection risk within shared communities, a two-level Bayesian hierarchical logistic regression was constructed. Village-level animal infection status was used as the primary exposure variable, while other covariates consisted of individual-level factors retained from the human multivariable logistic regression model. Bayesian hierarchical models were fitted using the brms package in R, employing weakly informative priors for regression coefficients and standard deviations (normal (0, 0.5)), and half-Cauchy priors (0.5) for village-level variance. Model convergence was assessed using Rhat statistics. To ensure convergence of the algorithm, we ran four chains with 2000 warm-up iterations each, followed by another 2000 sampling iterations, retaining a total of 8000 posterior samples. Rhat values close to 1.00 and effective sample sizes exceeding 500 were considered indicative of good convergence. Results were reported as medians with 95 % CI. We conducted three Bayesian hierarchical logistic regressions: Model 1 included village-level variables, Model 2 individual-level variables, and Model 3 (final model) both levels. Bayesian information criteria (Watanabe - Akaike information) evaluated predictive performance, with lower scores indicating better prediction accuracy.

3. Results

3.1. Human infection

Among the 1013 participants surveyed, 309 were infected with *C. sinensis*, resulting in a prevalence of 30.50 % (95 % CI: 27.68–33.44 %) (Table 1). The geometric mean EPG among infected individuals was 125 (95 % CI: 113–139).

At the village level, Nongdong recorded the highest prevalence (85/210; 40.48 %), followed by Bayin (73/185; 39.46 %) and Houzhai (62/202; 30.69 %). Infection was notably higher in males (247/513; 48.15 %), among individuals who had a history of consuming raw freshwater fish (228/418; 54.55 %),in the age group of 30–44 years old (58/125; 46.40 %), and among residents with a junior high school education (244/640; 38.13 %).

3.2. Risk factors for human *C. sinensis* infection

In univariate logistic regression analyses, village of residence, gender, awareness of clonorchiasis, history of consuming raw freshwater fish, frequency of raw fish consumption, frequency of raw fish preparation, use of separate cutting boards for raw and cooked foods, receipt of raw freshwater fish as a gift, and household animal ownership (cats, dogs, livestock, or poultry) were all significantly associated with infection (Table 2). In the multivariable logistic regression model, male was associated with a significantly higher risk of *C. sinensis* infection (adjusted odds ratio (aOR) = 7.53, 95 % CI: 2.76–22.27) compared with female sex. Individuals with a history of consuming raw freshwater fish was associated with a higher risk of *C. sinensis* infection (aOR = 6.91, 95 % CI: 1.69–33.25) compared with those who never consumed raw freshwater fish.

3.3. Animal infection

A total of 802 animals were surveyed, of which six were detected with *C. sinensis*, yielding an overall prevalence of 0.75 % (95 % CI: 0.28–1.62) (Table 3). Houzhai recorded the highest prevalence at 1.79 % (95 % CI: 0.22–6.30). The geometric mean EPG among five infected cats was 133 (95 % CI: 106–167), while the only infected dog had an EPG of 60. Apart from cats and dogs, none of the other surveyed animal species were found to be infected.

3.4. Risk factors for *C. sinensis* infection in animal hosts

Among the 169 dogs surveyed, the infection prevalence was 0.59 %

Table 1
Prevalence of *Clonorchis sinensis* infections in humans in Fusui County, Guangxi, 2024 ($n = 1013$).

Variable	No. examined	No. with *C. sinensis* infection	Prevalence % (95 % CI)	Geometric mean of EPG (95 % CI)
Villages				
Lianhao	210	50	23.81 (18.22–30.16)	78 (64–96)
Nongdong	210	85	40.48 (33.78–47.45)	214 (175–261)
Bayin	185	73	39.46 (32.37–46.90)	106 (84–133)
Xichang	206	39	18.93 (13.82–24.96)	130 (105–161)
Houzhai	202	62	30.69 (24.41–37.55)	104 (86–127)
Gender				
Female	500	62	12.40 (9.64–15.61)	93 (74–117)
Male	513	247	48.15 (43.75–52.57)	135 (120–151)
Have eaten raw freshwater fish				
No	595	81	13.61 (10.96–16.63)	92 (77–110)
Yes	418	228	54.55 (49.63–59.39)	139 (123–158)
Age/year				
≤ 14	33	1	3.03 (0.08–15.76)	72 (–)
15–29	32	6	18.75 (7.21–36.44)	145 (87–243)
30–44	125	58	46.40 (37.44–55.54)	117 (92–150)
45–59	403	126	31.27 (26.77–36.04)	119 (101–140)
≥ 60	420	118	28.10 (23.85–32.66)	136 (114–161)
Occupation				
Farmer	974	309	31.72 (28.81–34.75)	125 (113–139)
Student	35	0	–	–
Official	4	0	–	–
Education				
Illiteracy or semi-illiteracy	35	2	5.71 (0.70–19.16)	42 (14–122)
Primary school	313	59	18.85 (14.67–23.63)	134 (105–171)
Junior high school	640	244	38.13 (34.35–42.01)	124 (110–139)
Senior high school and above	25	4	16 (4.54–36.08)	169 (46–622)
Total	1013	309	30.50 (27.68–33.44)	125 (113–139)

Table 2
Univariate and multivariate logistic regression analysis of factors related to *Clonorchis sinensis* infection in population (*n* = 200).

Characteristics		No. examined	No. with *C. sinensis* (%)	Crude OR (95 % CI)	*P*	Adjusted OR (95 % CI)	*P*
Villages					< 0.05		
	Nongdong	40	18 (45)				
	Lianhao	40	28 (70)	2.85 (1.15–7.33)	< 0.05		
	Houzhai	40	19 (47.50)	1.11 (0.46–2.68)	0.82		
	Xichang	40	13 (32.50)	0.59 (0.23–1.45)	0.25		
	Bayin	40	25 (62.50)	2.04 (0.84–5.06)	0.12		
Gender					< 0.05		< 0.05
	Female	77	10 (12.99)				
	Male	123	93 (75.61)	20.77 (9.88–47.69)		7.53 (2.76–22.27)	
Age							
	15–29	5	1 (20.00)				
	30–44	29	19 (65.52)	7.60 (0.97–159.74)	0.09		
	45–59	72	37 (51.39)	4.23 (0.59–84.90)	0.21		
	≥ 60	94	46 (48.94)	3.83 (0.54–76.50)	0.24		
Education							
	Illiteracy or semi-illiteracy	8	2 (25.00)				
	Primary school	45	20 (44.44)	2.40 (0.49–17.61)	0.31		
	Junior high school	142	81 (57.04)	3.98 (0.88–27.83)	0.10		
	Senior high school and above	5	0 (0)				
Hearing of clonorchiasis					< 0.05		
	No	58	22 (37.93)				
	Yes	142	81 (57.04)	2.17 (1.17–4.11)			
Knowing transmission route					0.07		
	No	68	29 (42.65)				
	Yes	132	74 (56.06)	1.72 (0.95–3.12)			
Knowing harm					0.63		
	No	114	57 (50.00)				
	Yes	86	46 (53.49)	1.15 (0.66–2.02)			
Have eaten raw freshwater fish					< 0.05		<0.05
	No	55	5 (9.09)				
	Yes	145	98 (67.59)	20.85 (8.50–63.03)		6.91 (1.69–33.25)	
Frequency on consuming raw freshwater fish					< 0.05		
	No	92	25 (27.17)				
	< 5 times/year	55	38 (69.09)	5.99 (2.92–12.75)	< 0.05		
	5–10 times/year	37	27 (72.97)	7.24 (3.15–17.77)	< 0.05		
	> 10 times/year	16	13 (81.25)	11.61 (3.41–53.82)	< 0.05		
Frequency of making raw freshwater fish					< 0.05		
	Never	72	23 (31.94)				
	1–10 times/year	107	64 (59.81)	3.17 (1.71–6.02)	< 0.05		
	> 10 times/year	21	16 (76.19)	6.82 (2.36–22.97)	< 0.05		
Use separate cutting boards for raw and cooked foods					< 0.05		
	No	123	75 (60.98)				
	Yes	77	28 (36.36)	0.37 (0.20–0.65)			
Purchased raw freshwater fish from outside					0.12		
	No	98	45 (45.92)				
	Yes	102	58 (56.86)	1.55 (0.89–2.72)			
Gave raw freshwater fish to neighbors					0.90		
	No	158	81 (51.27)				
	Yes	42	22 (52.38)	1.05 (0.53–2.08)			
Received raw freshwater fish as a gift					< 0.05		
	No	137	63 (45.99)				
	Yes	63	40 (63.49)	2.04 (1.11–3.81)			
Whether to keep cat/dog/livestock/poultry					< 0.05		
	No	20	5 (25.00)				
	Yes	180	98 (54.44)	3.59 (1.33–11.40)			

Table 3

Prevalence of *Clonorchis sinensis* infections in animal in Fusui County, Guangxi, 2024 ($n = 802$).

Variable		No. examined	No. with *C. sinensis* infection	Prevalence % (95 % CI)	Geometric mean of EPG (95 % CI)
Villages					
	Lianhao	229	0	0	
	Nongdong	246	3	1.22 (0.25–3.52)	107 (59–193)
	Bayin	111	1	0.90 (0.02–4.92)	180 (–)
	Xichang	104	0	0	
	Houzhai	112	2	1.79 (0.22–6.30)	107 (86–134)
Animal gender					
	Male	503	3	0.60 (0.12–1.73)	107 (59–193)
	Female	298	3	1.01 (0.21–2.91)	128 (89–183)
Animal species					
	Cat	53	5	9.43 (3.13–20.66)	133 (106–167)
	Dog	169	1	0.59 (0.01–3.25)	60 (–)
	Chicken	151	0	0	0
	Duck	206	0	0	0
	Goose	27	0	0	0
	Cattle	39	0	0	0
	Sheep	103	0	0	0
	Pig	54	0	0	0
Total		802	6	0.75 (0.28–1.62)	117 (85–161)

(95 % CI: 0.01–3.25), whereas among the 53 cats, it was 9.43 % (95 % CI: 3.13–20.66). Table 4 summarized the results of univariate and multivariable logistic regression analyses of host factors in dogs and cats. In the multivariable model, only host species remained significantly associated with infection: compared to dogs, cats were associated with a higher risk of *C. sinensis* infection (aOR = 17.50, 95 % CI: 2.74–339.59).

3.5. *Association between animal and human infections*

We fitted three Bayesian multilevel logistic regression models to assess the impact of village-level animal infection on human *C. sinensis* infection risk. Posterior regression coefficients, posterior odds ratios with 95 % CI, random-effect standard deviations (SD), Rhat, and effective sample sizes (ESS) are presented in Table 5. The reported estimates for both regression coefficients and variance components correspond to the means of their posterior distributions. Model 2 included only individual-level covariates selected from the prior multivariate analyses, whereas Models 1 and 3 additionally incorporated a binary indicator for whether any animal in the respondent's village tested positive for *C. sinensis*.

In Model 2 (without the animal infection variable), the village level random effect standard deviation was 0.65 (95 % CI: 0.30–1.36), indicating substantial clustering of infection at the village level. After adding the animal-infection indicator, the village-level standard deviation decreased markedly (Model 1: SD = 0.24; Model 3: SD = 0.18), suggesting that animal infections drive much of the between village heterogeneity. Among the three models, Model 3 which includes both individual-level covariates and village level animal infection, had the lowest WAIC (968.90), and was therefore selected for inference.

In Model 3, village-level animal infection was associated with a 2.37-fold increase in the odds of human infection (aOR = 2.37, 95 % CI: 1.23–3.58). Male sex was associated with higher risk compared to female (aOR = 3.46, 95 % CI: 2.44–4.98), and a history of consuming raw freshwater fish resulted in a 5.27-fold increase in odds relative to never consuming raw fish (aOR = 5.27, 95 % CI: 3.70–7.48).

We evaluated convergence for Model 3 through trace plot diagnostics. These plots exhibited well-behaved stationary distributions fluctuating around a central equilibrium, consistent with reliable MCMC sampling (Fig. 1). Rhat for all parameters was 1.00, indicating good convergence (Table 5).

4. Discussion

Guided by the One Health concept, this study comprehensively assessed *C. sinensis* infection among humans and animal hosts within shared communities in Fusui County, Guangxi. Although the human infection prevalence observed in 2024 (30.5 %) declined compared to the previously reported rate (40.3 %) from 2002 [25], it remains indicative of hyperendemic transmission [26]. Thus, clonorchiasis continues to pose a significant public health challenge in the region.

Among eight surveyed animal species, only dogs (0.59 %) and cats (9.43 %) were detected with *C. sinensis*, with a significantly higher prevalence in cats than in dogs. This difference is probably related to differences in behavior, diet, or husbandry practices. For example, cats have free-roaming habits and could prey on fish [27]. This study found that most cats were free-roaming, whereas the majority of dogs were kept in cages. Thus, cats had more chances to ingest raw freshwater fish. The field survey found no infection in poultry (chickens, ducks, or geese), consistent with Shao et al.'s laboratory findings [16], indicating that poultry do not play a significant role in this study area, and even do not serve as suitable hosts, in the transmission cycle of *C. sinensis*. Among

Table 4

Univariate and multivariate logistic regression analysis of factors related to *Clonorchis sinensis* infection in cats and dogs.

Characteristics		No. examined	No. with *C. sinensis* (%)	Crude OR (95 % CI)	*P*	Adjusted OR (95 % CI)	*P*
Age							
	< 1 year	148	4 (2.7)				
	1- < 3 years	64	2 (3.12)	1.16 (0.16–6.11)	0.86		
	3–10 years	10	0 (0)		0.99		
Animal gender							
	Male	146	3 (2.05)				
	Female	76	3 (3.95)	1.96 (0.35–10.81)	0.42		
Animal species							
	Dog	169	1 (0.59)				
	Cat	53	5 (9.43)	17.50 (2.74–339.59)	< 0.05	17.50 (2.74–339.59)	< 0.05

Table 5
Bayesian logistic regression model of the impact of village animal infection on human infection.

Parameter	Model 1				Model 2				Model 3			
	Estimate (SE)	OR (95 % CI)	Rhat	ESS	Estimate (SE)	OR (95 % CI)	Rhat	ESS	Estimate (SE)	OR (95 % CI)	Rhat	ESS
Fixed effects												
Intercept	−1.19 (0.23)		1.00	2324	−2.34 (0.34)		1.00	2334	−2.95 (0.25)		1.00	3834
Animal infection in the village (Yes)	0.59 (0.27)	1.80 (0.97–2.81)	1.00	2548					0.86 (0.26)	2.37 (1.23–3.58)	1.00	3101
Gender (Male)					1.23 (0.19)	3.42 (2.41–4.96)	1.00	4787	1.24 (0.18)	3.46 (2.44–4.98)	1.00	7012
Have eaten raw freshwater fish (Yes)					1.69 (0.18)	5.40 (3.77–7.70)	1.00	4926	1.63 (0.18)	5.27 (3.70–7.48)	1.00	6255
Random effect	Estimate (SE)	95 % CI	Rhat	ESS	Estimate (SE)	95 % CI	Rhat	ESS	Estimate (SE)	95 % CI	Rhat	ESS
Village intercept SD	0.24 (0.20)	(0.02–0.74)	1.00	1565	0.65 (0.28)	(0.30–1.36)	1.00	2398	0.18 (0.15)	(0.01–0.67)	1.00	1883
Model fit indicator												
WAIC		1220.80				971.70				968.90		

ESS, effective sample sizes.
WAIC, Watanabe-Akaike information criterion.

the surveyed livestock, no infections were detected in cattle or sheep. As strict herbivores that typically do not consume fish, it is highly unlikely that they would acquire the infection via the foodborne route. Notably, no infections were detected in pigs despite their omnivorous diet, a finding that differs from the report by Lin's report [13]. Only a few households raised pigs in the surveyed villages, and probably the absence of using raw freshwater fish as swine feed prevented infection in pigs. These results underscore that control strategies for *C. sinensis* need be guided by local epidemiological evidence rather than by applying a generic host list. In summary, this survey delineates a clear picture of local transmission in animal reservoirs: it is driven by cats and dogs as core reservoir hosts, whereas poultry (chickens, ducks, or geese) and major livestock (pigs, cattle, and sheep) appear not to participate in the cycle.

Our Bayesian multilevel logistic regression models identified several key determinants of *C. sinensis* infection risk. At the individual level, the consumption of raw freshwater fish remains the primary risk factor for *C. sinensis* infection [28,29]. The odds of infection for consumers are significantly increased (aOR = 5.27) higher than for non-consumers, and this risk further escalates with the frequency of consumption. This behavior is not a sporadic personal choice but a deeply ingrained cultural tradition, continually reinforced through frequent social activities. For instance, the custom of “receiving raw freshwater fish as a gift” highlights its social dimension, elevating it beyond a mere dietary habit to a socially reinforced cultural symbol that leads to continuous and widespread population exposure. Furthermore, males are at a significantly higher risk of infection than females (aOR = 3.46). This disparity is likely explained by the fact that, compared to women, men may more frequently participate in communal dining, banquets, and other social gatherings where raw freshwater fish is often served as an essential dish [30,31]. At the village level, the presence of infected animals emerged as a significant driver of between-village heterogeneity in human infection rates: After controlling for individual-level factors, the presence of infected animals in a village was associated with a 2.38-fold increase risk of human infection. The sustained environmental contamination from animal reservoirs' feces facilitates ongoing parasite transmission cycles through snail intermediate hosts and freshwater fish [32–34], thus indirectly raising human infection risk. These findings highlight the potential benefits of broadening clonorchiasis prevention strategies beyond a purely human - centric approach. Integrated interventions might therefore include animal management (particularly cats and dogs), improved fecal waste handling, regular surveillance of intermediate snail and fish hosts, and community health education campaigns focusing on promoting safe dietary practices and raising awareness of parasite transmission pathways. Such comprehensive strategies may contribute substantially to reducing environmental transmission and overall human infection risk.

4.1. Limitations

Our study has several limitations. First, the analysis was based on data from only five villages, which may limit the universality and external validity of the findings, especially given the potential heterogeneity across different communities. To enhance the robustness of the results, we employed a Bayesian statistical approach that incorporates prior information to address uncertainty associated with small sample sizes, thereby yielding more stable estimates. Future research should expand the study area to include a larger number of villages, which would allow for further validation of the findings and improve their representativeness and applicability. Second, because the numbers of geese, cattle, cats and pigs were limited in the sampled villages, the achieved sample sizes for these species did not meet the planned targets, which may have reduced the precision. Future studies are expected to increase the number of these species in villages raising them. Third, the cross-sectional design precludes establishing causality or temporal sequencing between animal infections and human disease outcomes; future prospective cohort studies or molecular tracing analyses are required to clarify the transmission dynamics. Especially, combining molecular genotyping analysis of adult parasites or eggs in humans, cats, and dogs could validate the existence of cross-host transmission chains, which would significantly strengthen the persuasiveness of the transmission pathway from animals to the environment to humans. Fourth, surveys on intermediate hosts (freshwater snails and fish) were not included, which need to be integrated in future to fully characterize transmission pathways. Finally, this study employed only two Kato-Katz smears from single sample for examination, which may underestimate the infection, especially in animals with low-intensity infections. Thus, molecular diagnostic techniques (e.g., PCR) could be considered to include in future work.

5. Conclusion

Under the One Health framework, our study demonstrates that poultry (chickens, ducks, geese) and livestock (pigs, cattle, sheep) do not appear participate in local transmission, and the presence of infected animal hosts (particularly cats) within shared community environments significantly associated with an increased risk of human *C. sinensis* infection (aOR = 2.37). This evidence emphasizes the critical

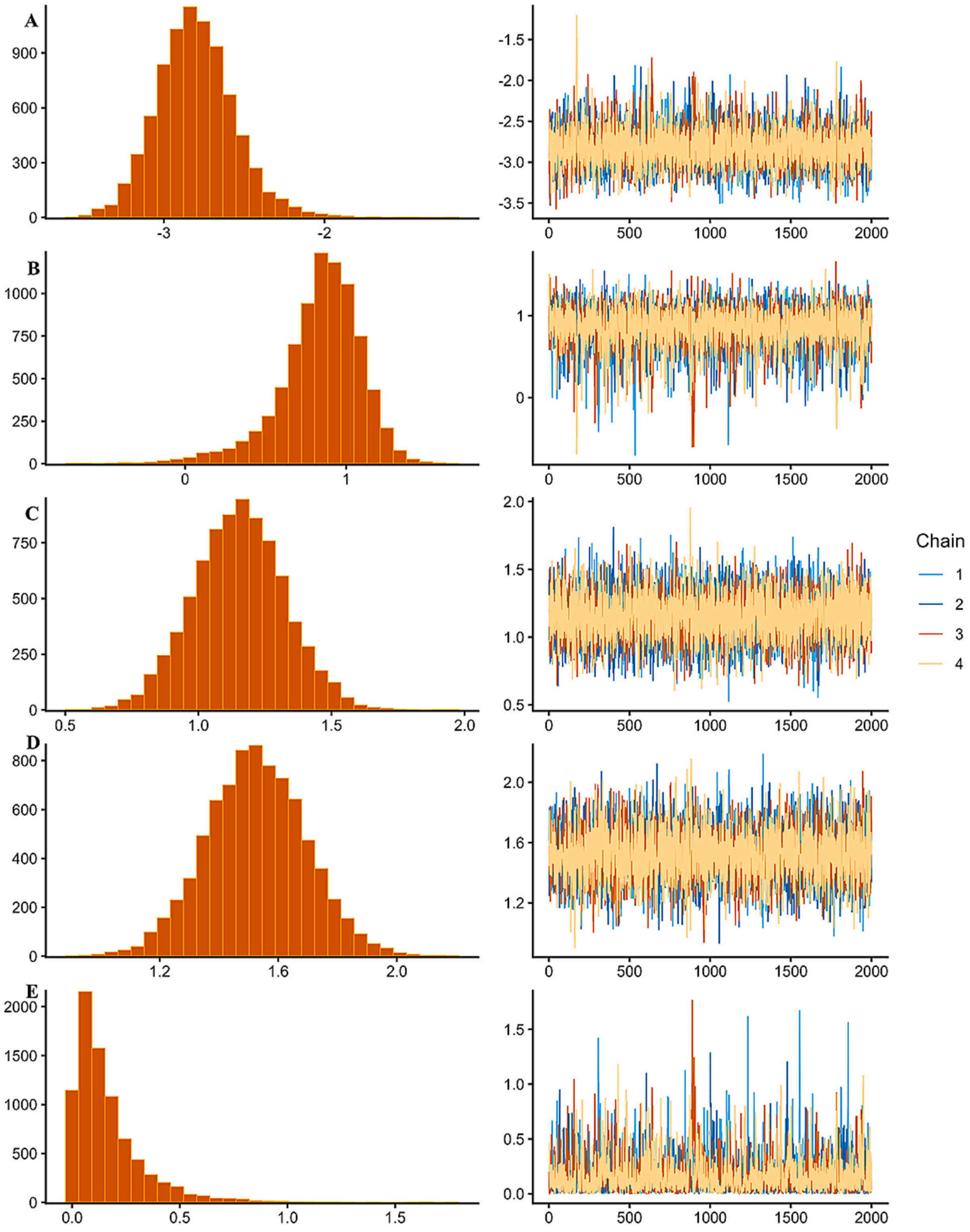

Fig. 1. Diagnostic plots for the Bayesian hierarchical model (Model 3).
The figure displays the posterior distribution histograms (left panels) and the MCMC trace plots for the four chains (right panels), indicating successful model convergence. The panels from top to bottom represent the parameters for: (A) the model intercept, (B) animal infection in the village, (C) gender, (D) history of eating raw freshwater fish, and (E) the standard deviation of the village-level random effect.

importance of transitioning from isolated human-focused interventions to an integrated, multisectoral strategy combining animal health management, environmental sanitation, continuous host surveillance, and community education, thereby enabling sustainable and effective clonorchiasis control.

CRediT authorship contribution statement

Xiao-Ping Han: Writing – review & editing, Writing – original draft, Visualization, Validation, Software, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Ting-Jun Zhu:** Supervision, Resources, Project administration, Methodology, Investigation. **Liu Liu:** Methodology, Investigation. **Chang-Hai Zhou:** Methodology, Investigation. **Zhi-Yue Huang:** Methodology, Investigation. **Sheng-Jun Wei:** Methodology, Investigation. **Lu-Yuan Zhao:** Methodology, Investigation. **Jun Meng:** Supervision, Conceptualization. **Shi-Zhu Li:** Supervision, Conceptualization. **Men-Bao Qian:** Writing – review & editing, Supervision, Resources, Project administration, Methodology, Funding acquisition, Conceptualization.

Ethical approval and consent to participate

The study was approved by the Ethical Review Committee of National Institute of Parasitic Diseases, Chinese Center for Disease Control and Prevention (Chinese Center for Tropical Disease Research) (ref: 2024–006). Written informed consent was obtained from all participants or their guardians for the children.

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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 paper.

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Data availability

Data are not publicly available but are available on reasonable request after reviewed by the National Institute of Parasitic Diseases, Chinese Center for Disease Control and Prevention.

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