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Foodborne Human Fascioliasis in Pakistan: A Review

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Abstract

Human fascioliasis is a neglected parasitic disease affecting millions of people worldwide. Understanding geographical prevalence and environmental contamination is critical for effective risk assessment, prioritization, and mitigation of biological hazards. This literature review aims to synthesize the dynamics of foodborne transmission, the geographical distribution, and the prevalence of human fascioliasis in Pakistan. The included studies (n=13), all from the Punjab or Khyber Pakhtunkhwa provinces, investigated *Fasciola* spp. in humans (n=6), vegetables (n=4), and water (n=3). The pooled prevalence in humans based on stool microscopy was 0.73% (95% CI: 0.61% - 0.88%), and up to 23% of the population was seropositive, depicting a high exposure to this parasite. The burden of fascioliasis in Pakistan was estimated to be at least 36,000 disability adjusted life years (DALYs). The prevalence varied based on geographic, demographic, diagnostic, and socioeconomic factors. Vegetable contamination at 1.92% (95% CI: 1.26-2.91%) was mostly higher in leafy vegetables. Water contamination with parasitic larvae or eggs varied across sources, including 1.6% in tap water, 10% in tube wells, 8% in open wells, 6% in a pond, and 10.7% in water drains. Overall, the literature indicated that fascioliasis is endemic in Pakistan, with disparities in geographical prevalence and research focus. The analysis presented here will move a step closer to guiding researchers, public health professionals, and policymakers in evidence-based decision-making in resource allocation, surveillance, and targeted control measures to reduce fascioliasis burden in Pakistan.

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Introduction

Liver flukes (*e.g.*, *Fasciola hepatica* and *F. gigantica*) are responsible for parasitism in both humans and domestic animals, particularly ruminants [1–3]. Hosts become infected by ingesting the larvae (metacercariae) through the consumption of contaminated water, vegetables, and aquatic plants (Fig. 1) [4]. Fascioliasis is declared as a neglected tropical disease by the WHO under the classification of food-borne trematode [1], and the FAO ranks *Fasciola* spp. 12th on the foodborne parasite priority list [4, 5]. Once ingested, the larvae migrate through the liver and bile ducts, causing tissue damage and inflammation. Immunologically, the larval migration triggers a complex immune cascade [2]. Initially, the host mounts an innate inflammatory response characterized by the recruitment of neutrophils and macrophages to the site of tissue damage [3]. However, *Fasciola* spp. have evolved sophisticated immunomodulatory mechanisms, such as the secretion of cathepsin L proteases, which cleave host immunoglobulins and suppress the Th1-cell-mediated response, favoring a Th2-polarized environment that allows for parasite persistence and chronic infection [6, 7].

The global prevalence of human fascioliasis is estimated to range from 2.4 to 17 million individuals, with millions at risk of transmission across more than 70 countries [1,4,5]. Rosas-Hostos Infantes et al. [8] reported a pooled prevalence of 4.5% in 15 countries, including Bolivia (21%), Peru (11%), and Egypt (6%). In Pakistan, the prevalence has been reported to range from 0.6% to 27% [9]. The majority of studies in Pakistan are on *Fasciola* spp. in ruminants [2,3,9]. However, human fascioliasis is emerging due to changing climate dynamics [10]. Foodborne parasites cause morbidity and mortality in humans and pose a significant public health burden globally. The global burden of four main foodborne trematodes was estimated to be up to two million disability-adjusted life-years (DALYs) [11,12]. In Pakistan, these figures are expected to be different due to changing climate, socioeconomic, and cultural factors, which create ideal conditions for parasitic endemicity. Moreover, these foodborne parasites may persist without clinical signs in the host and cause nutritional deficiencies, thereby making the host more prone to other communicable and non-communicable diseases.

There are two literature reviews previously published on human fascioliasis, which included 2 to 3 original studies from Pakistan [8,9]. There is no review with a larger number of human studies from Pakistan. Meanwhile, foodborne transmission remains the primary route of human fascioliasis, and no comprehensive study has reported nationwide contamination rates in food and water. Therefore, this literature review analyses the prevalence of human fascioliasis, associated risk factors, and their contamination in the food chain. It will also provide expert recommendations to guide future research initiatives, evidence-based control measures, and efficient policy development for mitigating the burden of foodborne parasitic diseases in the country.

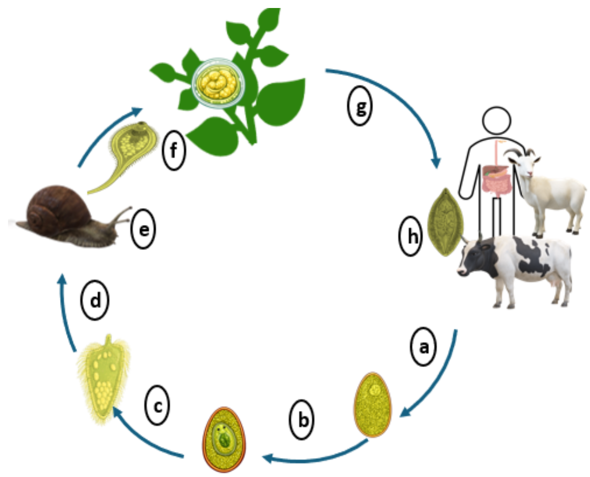

Fig. 1 Lifecycle of *Fasciola* spp.: **a)** Unembryonated parasite eggs from definitive host (ruminants) or accidental host (human) are excreted in feces. **b)** Eggs embryonate in the water. **c)** First-stage larvae (miracidia) are hatched and, **d)** lodge in an intermediate host (water snail). **e)** After going through further larval stages of sporocysts and radiae, they become cercariae and swim freely in water. **f)** Cercariae encyst on water vegetation to become metacercariae. **g)** Hosts are exposed to this infectious stage through ingestion of contaminated water or plants. **h)** Metacercariae excyst in the duodenum and migrate through the intestinal wall into the peritoneal cavity, passing liver parenchyma and finally into the biliary duct, maturing into an adult fluke.

Methods

The PRISMA methodology was employed for literature search and data extraction (Fig. 2). In the first step, the literature was searched in PubMed, Scopus, and Web of Science. A different combination of keywords was used, including (Foodborne OR zoonotic) AND (trematode* OR fluke* OR fasciola) AND (Pakistan OR South Asia OR Punjab OR Sindh OR Khyber Pakhtunkhwa OR Balochistan OR Sarhad OR Gilgit OR Kashmir).

Inclusion criteria were: i) studies on *Fasciola* spp. in Pakistan (human/environment), ii) original research, iii) publications in the English language

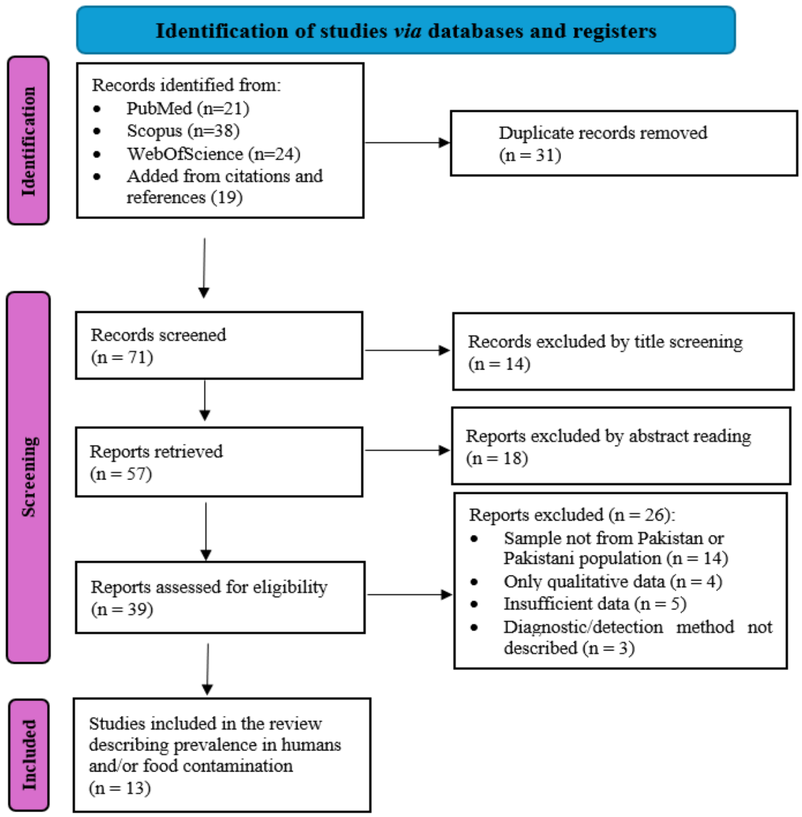

Fig. 2 PRISMA flow diagram of study selection process.

(peer-reviewed articles, theses, reports), and iv) published during 2000–2026. The studies were excluded if: i) primary data were missing (*e.g.*, reviews without new findings), ii) data were not from Pakistan, iii) only quantitative results were documented, iv) case reports (sample size <5), and v) diagnostic or detection methods were not described. The results are presented in tables (Tables 1 and 2). Prevalence and positivity rate were calculated as the proportion of positive samples to the total tested samples/population. The pooled prevalence and positivity were obtained by combining the number of cases (or positives) and the population (sample) size across the studies. A 95% confidence interval (CI) was estimated for the combined proportion using Clopper–Pearson (for population prevalence) or Wilson score (for environmental sample positivity) methods.

As an indicator of disease burden, DALYs are calculated as the sum of years of life lost (YLL) and years lived with disability (YLD) [13]. As there is negligible YLL for *Fasciola* spp., we set DALYs=YLD [11]. To estimate the effect of fascioliasis, YLDs were estimated as *eq. 1*:

YLD = Prevalence × Disability Weight × Duration × Population …. *eq. 1*

where prevalence was taken as the pooled prevalence, disability weight was taken as 0.02, for an average duration of one year, and a population of 250 million Pakistanis.

Results

Six studies have reported human fascioliasis in Pakistan between the years 2003 and 2016 (Table 1). Four studies were conducted in Punjab and Islamabad Capital Territory (ICT), and two were reported from Khyber Pakhtunkhwa (KPK). Regarding contamination of food and water, three studies were conducted in Punjab and four in KPK. There was no data available from other parts of Pakistan.

Human fascioliasis in Pakistan

In the early 2000s, the seroprevalence in Punjab was reported as 4.67% (2.28% – 7.05%) using the indirect hemagglutination (IHA) method [14]. Later, an enzyme-linked immunosorbent assay (ELISA) study reported a seropositivity of 23.8% (95% CI: 20.2% – 27.4%) for *Fasciola* IgG antibodies in the ICT and Punjab in 2016 [15]. The prevalence based on coprological microscopic examination was 0.73% (95% CI: 0.61–0.88%, 111 positives out of 15124

Table 1 Prevalence of *Fasciola* sp. in humans in Pakistan**.**

Area	Duration	Method	Population	Prevalence *	Influencing variables	Study
Total	April 2003- March 2005	IHA of serum	Male, female; all ages (0-10, 11-20, 21-30, >30)	4.67% (14/300; 2.28% – 7.05%)	Geographical area and age	[14]
Kamonki (PNB)				8% (4/50)		
Muridke (PNB)				6% (3/50)		
Shandra (PNB)				6% (3/50)		
Kasur (PNB)				4% (2/50)		
Gujranwala (PNB)				2% (1/50)		
Sheikhupura (PNB)				2% (1/50)		
Total	May 2014 - August 2016	ELISA of serum IgG	Male, female; all ages	23.8% (130/546; 20.24% – 27.38%)	City, locality (rural/urban), gender, age, family size, hygiene practice, season, medication, socioeconomic status, drinking untreated water	[15]
Bharakahu (ICT)				42.2% (32/74)		
Golra Mor (ICT)				14.9% (14/94)		
Islamabad (ICT)				7.4% (8/108)		
Rawalpindi (PNB)				33.3% (16/48)		
Faisalabad (PNB)				34.5% (20/58)		
Gujranwala (PNB)				6.8% (6/88)		
Muzaffargarh (PNB)				44.7% (34/76)		
Total	April 2003- April 2005	Microscopy of stool samples	Male, female; all ages (0-10, 11-20, 21-30, >30)	1.18% (85/7200; 0.93% – 1.43%)	Geographical area, age, season/month	[16]
Kamonki (PNB)				1.75% (21/1200)		
Muridke (PNB)				1.3% (16/1200)		
Shandra (PNB)				1.58% (19/1200)		
Kasur (PNB)				0.92% (11/1200)		
Gujranwala (PNB)				0.83% (10/1200)		
Sheikhupura (PNB)				0.67% (8/1200)		
Rural Lahore (PNB)	April 2003 - March 2005	Microscopy of stool samples	Male, female; all ages (0-10, 11-20, 21-30, >30)	0.29% (21/7200)	Season, month, location, age	[17]
Mardan district (KPK)	September - May	Microscopy of stool samples	Male, female, all ages	0.74% (4/540)	Geographical area, age	[18]
Malakand (KPK)	February - June 2016	Microscopy of stool samples	Male children aged 14±3.05 years	0.54% (1/184)	NA	[19]

PNB = Punjab, KPK = Khyber-Pakhtunkhwa, IHA, ELISA, CI = confidence interval of 95%, NA = no variables studied or no variables with significant (p<0.05) effect. *prevalence % (positive / total; 95% CI: range)

tested samples). In KPK province, the prevalence was 0.69% (95% CI, 0.22%–1.60%; 5/724), with 0.5% in children from Malakand [19] and 0.74% (urban area 0.5%, rural area 0.9%) in Mardan [18]. Whereas, in Punjab, the prevalence was documented as 0.74% (95% CI, 0.60% – 0.89%, 106/14400), with 0.3% in rural Lahore [17] and 1.2% (0.9% – 1.4%) in other neighboring districts [16]. Considering a 0.73% prevalence (approximately 1.8 million infections / 250 million population), the burden of fascioliasis as YLD (DALYs) in Pakistan was estimated as 36,000.

Risk factors influencing prevalence

The prevalence varies by city, locality (rural/urban), gender, age, family size, hygiene practices, season, antiparasitic medication use, socioeconomic status, drinking untreated water, and eating raw/uncooked vegetables (Table 1). A higher prevalence rate was associated with age: 1.6% in 11-20 years, 1.2% in 0–10 years, 0.5% in 21–30 years, and the lowest in the above 30 age group [16]. Gender-based results showed that 6.6% of females and 17.2% of males were positive for *Fasciola* antibodies (odds ratio 3.6) [15]. Monthly prevalence showed two peaks, one in August (4.7%) and the other in January (2.2%) [16].

Diagnostic methods

Four studies used microscopic examination of parasite eggs excreted in feces for diagnosing *Fasciola* spp. [16–19]. However, studies have used ELISA and IHA to detect antigens or antibodies in human and animal blood [14, 15]. Detection of this

Table 2 Food and water contamination by *Fasciola* spp. in Pakistan.

Area	Sample	Method*	Positivity %	Details	Study
Lahore (PNB)	Vegetables (beet, cabbage, carrot, chili, coriander, cucumber, lettuce, mint, radish, tomato)	SD and FL; Microscopy	1.6% (8/500)	Overall parasite contamination rate was 31.6% (156/500). 1 *Fasciola* was positive in each vegetable except in chili and radish.	[20]
Lahore (PNB)	Vegetables (cucumber, cabbage, lettuce)	SD and FL; Microscope	2% (3/150)	Overall parasite contamination rate was 14% (21/150). Study conducted from January to April. One *Fasciola* was found in each vegetable.	[21]
Narowal (PNB)	Vegetables (coriander, spinach, mint, chili, and carrot)	SD and FL; Microscope	2.1% (3/145)	The overall parasite contamination rate was 47.6%. Study conducted from December to March. *Fasciola* positive 2 in mint and 1 in coriander.	[22]
Mardan (KPK)	Vegetables (lettuce, mint, coriander, cabbage, cucumber, chili, carrot)	Microscopy	2.3% (7/300)	Overall parasite contamination rate was 30.7% (92/300). Study conducted from October to April.	[23]
Karak, Kohat, Hangu (KPK)	Water (tap water, ponds, drain water)	SD; Microscopy	Tap water 2% (3/150), pond water 6% (9/150), and drain water 6.7% (10/150)	Overall contamination in all sources: 4.8% (22/450)	[24]
Dera Ismail Khan (KPK)	Water (tap water, ponds, drain water)	SD; Microscopy	Tap water 1% (1/100), pond water 6% (3/50), and drain water 12% (6/50)	Overall contamination in all sources: 5% (10/200)	[25]
Nowshehra (KPK)	Water (tubewell, open well, tap water, drain water)	PCR, primer: Fas-F, 5'AGTGAT TACCCGC TGAACT3	Tap water 1.66 % (1/60), tube well 10% (4/40), open well water 8 % (8/100), drain 16% (16/100)	The overall prevalence across different water sources was 9.66 % (29/300).	[26]

*Sample preparation and detection method; SD = Sedimentation, FL= Floatation

parasite (eggs, cercariae, metacercariae) in food and water samples is primarily performed by microscopic examination of samples prepared using flotation or sedimentation methods [20–25]. Only a single study reported using PCR to detect *Fasciola* spp. DNA in the water samples [26].

Food and water contamination

The contamination of water and vegetables by this parasite is shown in Table 2. Overall, vegetable contamination was 1.92% (95% CI: 1.26-2.91%), with 1.2% in Mardan [23], 1.6 to 2% in Lahore [20,21], and 2.1% (4% coriander and 9.5% mint) in Narowal [22]. Water contamination with parasitic larvae or eggs varied across sources, including 1.6% in tap water, 10% tube wells, 8% open wells, 6% pond, and 10.67% in drain water [24–26].

Discussion

The results showed a substantial variation in the prevalence of human fascioliasis and food and water contamination across various host and environmental factors in the country. Overall, a low number of studies depict the understudied status of human fascioliasis in Pakistan. The variation in prevalence across diagnostic methods, including coprological detection (up to 1.8%) and ELISA (23.8%), highlights diagnostic limitations and a gap in our understanding of the disease, known as the 'immunological window' [3]. In general, stool parasitological examination is considered a cheap and simple solution for rapid diagnosis and mass screening; however, it has low sensitivity in cases of low or chronic infestation, potentially leading to false-negative results and underestimation of infestations [2, 12]. On the other hand, serological detection methods detect exposure but cannot distinguish between present or past parasitism [15]. Therefore, it can be assumed that the true prevalence lies between these two figures. Future investigations should adopt antigen-specific or molecular methods to more accurately distinguish active from resolved infestations. The lower prevalence of fascioliasis in Pakistan than in other developing countries [8] may be associated with various factors, including eating habits [15]. For example, consumption of raw aquatic plants is uncommon in the country. Moreover, a previous study on the knowledge, attitudes, and practices related to fascioliasis among the human population in Punjab reported a 57% awareness of fascioliasis infection among respondents [27]. It can be assumed that if the population is aware of the risk, they will take precautionary measures to avoid the hazard [12, 28].

The two seasonal peaks of summer (July-August) and winter (January-February) are associated with the post-monsoon humidity and the irrigation cycles of winter [10, 15–17]. These activities allow a higher snail population and water contact with the food. Overall, the high infection rate was recorded in summer as compared to the winter season because summer provided more ideal conditions for these parasitic transmissions. Similar parasitism results were reported in the animal population as well [3, 9, 10]. The highest infestation rates among teenagers could be associated with behavioral exposure to the parasite. They are highly likely to be involved with high-exposure activities, such as swimming in open waters, harvesting fodder, or consuming raw vegetables in rural settings [14–19]. Because the number of positive samples was low, many studies did not detect a significant association between gender and the risk of parasitism. However, few studies in the literature have documented a slightly higher prevalence of fascioliasis in females [8, 15, 29]. Lower socioeconomic status and rural residence among individuals are also associated with higher rates of fascioliasis [14,16,18]. Socioeconomic status has been associated with access to healthcare, education, and a hygienic lifestyle; as the lower status limits access to these facilities, there is a higher risk of infections [15]. Regarding the higher rates of infestations in rural and agricultural environments, communities are often far from health care, have limited access to clean water, have poor personal and environmental hygiene, and are in close contact with intermediate and reservoir hosts, which maintain infection [10, 12]. Vegetable contamination (1.9%) is linked to the contaminated irrigation water. The true risk can be higher if infectious larval stages (metacercariae) are present in the water and contaminate the vegetables. Afshan et al. [15] reported eating raw vegetables as a risk factor for fascioliasis in the Pakistani population. However, this risk is generally considered low because, culturally, raw freshwater plants are not eaten in large quantities in the country. However, the risk from drinking water cannot be ignored, as various water sources, including those used for drinking, were contaminated [24–26]. Therefore, it is recommended to use clean water for drinking and irrigation operations [4, 28, 30]. *Fasciola* spp. are killed by cooking food at 60°C for several minutes and by boiling or filtering water. As this parasite is cold-resistant, chilling is not recommended [4].

The geographic skew in results, with no prevalence reports from Azad Jammu and Kashmir, Gilgit-Baltistan, Sindh, and Balochistan, reflects a lack of research focus in these areas rather than a true absence of disease [31–34]. Moreover, the most recent human fascioliasis data reported in the published studies are at least 10 years old; therefore, the current status of the disease is not accurately known. Meanwhile, other trematodes, such as *Schistosoma* sp. and *Opisthorchis* sp., are not considered here because of their rare importance in foodborne transmissions in Pakistan. However, research is also needed to assess the real risk posed by these trematodes. A high YLD (36,000) showed a significant burden of human fascioliasis. These figures are similar to previous studies; for example, Furst et al. [35] reported global YLD due to foodborne fascioliasis as 47,070 (for 2.6 million infections), and Torgerson et al. [11] reported it as 90,041 (ranging from 58,050 to 209,097). However, many other studies have shown that the real prevalence of fascioliasis could be even higher [12, 36]. Nevertheless, the YLD estimate has wide uncertainty because it relies on pooled prevalence estimates from a small number of studies, a uniform infestation duration of one year, a generalized disability weight, and does not account for the population's age-structured data. Therefore, future epidemiological studies are needed to more accurately model these estimates.

Another increasingly urgent dimension of fascioliasis is the growing resistance to available antiparasitic drugs. Triclabendazole remains the main livestock control drug and the only licensed or WHO-recommended drug for human fascioliasis [37]. However, zoonotic transmission of triclabendazole-resistant human fascioliasis has raised alarms [38]. Despite these concerns, a pronounced research gap exists on this subject in Pakistan. Without surveillance to fill this critical gap, the country risks silently harboring and amplifying antiparasitic resistance that could undermine control efforts on a far wider scale.

Overall, these findings underscore the need for a One Health strategy integrating control of *Fasciola* spp. in livestock and lymnaeid reservoirs, monitoring the safety of leafy and ground vegetables, and ensuring treatment of drinking and irrigation water in endemic areas. However, the absence of a truly homogeneous data representation across Pakistan remains a primary barrier in evidence-based policymaking. Moving forward, targeted surveillance using harmonized diagnostics in currently underexplored provinces is needed to quantify the true foodborne burden of fascioliasis and to prioritize resource allocations for control and prevention.

Conclusion

This review consolidates the current body of evidence on the foodborne burden of fascioliasis across Pakistan. The literature review revealed that human fascioliasis is an underestimated food and waterborne zoonosis. Only six studies have been conducted in northern and north-eastern parts of Pakistan, leaving large geographic blind spots of parasitism. Reported prevalence varies substantially across diagnostic methods, and no studies have used molecular or genomic methods to characterize this parasitism in humans. Contaminated water has also contaminated vegetables. These findings highlight the need for urgent, geographically representative sampling across Pakistan and for integrating the One Health approach, which targets livestock reservoirs, food safety, water quality, and overall sanitation, to mitigate foodborne fascioliasis nationwide.

Conflict of interest

The authors declare no conflicts of interest.

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