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Original papers

The seasonal incidence of parasitic helminth infection among the walking catfish, *Clarias batrachus* of Tripura, India

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ABSTRACT. In the present study, a total of 868 walking catfish, *Clarias batrachus,* were collected during the period of April, 2012 to March, 2015, to evaluate the prevalence of helminth parasites from different parts of Tripura, Northeast India. Of these, 606 fish were found to be infected with one genus of trematode viz., *Astiotrema*, two genera of cestode viz., *Lytocestus* and *Djombangia* and one genus of nematode parasite viz., *Anisakis*. Of the three groups of parasites recovered, the major group infecting the host were found to be cestodes, followed by nematodes and trematodes. Seasonal studies show that the overall prevalence of the collected helminth parasites was highest during the post-monsoon season, followed by the monsoon and pre-monsoon seasons.

Key words: *Clarias batrachus*, helminth, prevalence

Introduction

India is one of the 17 mega-diversity countries [1] which host as many as 55 families of freshwater fish [2]. In the Indian subcontinent, fish have been extensively used as food for human consumption, and this use contributes substantially to the economy of the region. It has been estimated that 10 million tons of fish are required to meet the annual demand for fish protein in India, compared to the actual production of only 3.5 million tons [3]. Catfish comprise a major group in the fish fauna and many of them serve as a food source, having high nutritive value. It has been reported that endoparasitic helminths with an indirect life cycle parasitize one or more definitive hosts belonging to the family Bagridae, Heteropneustidae, Schibeidae, Mastacembelidae and Clariidae leading to heavy economic losses [4–6].

Several workers have studied the helminth fauna of piscine hosts and have described several new species from India [7–17]. However, only limited information is available about the fish parasites of northeast India, and is mostly restricted to Meghalaya, Arunachal Pradesh and Assam [18–22]. Except for two preliminary records, no literature is available concerning helminth diversity among the various edible and economically important fishes of Tripura, Northeast India [23–24]. Dogiel et al. [25] state that seasonal environmental changes in water such as temperature, pH and conductivity affect the occurrence of parasites in aquatic hosts.

The present study examines the prevalence, abundance and mean intensity of helminth infections in the walking catfish, *Clarias batrachus*, for a period of three years extending from April, 2012 to March, 2015.

Materials and Methods

The sampling sites (Dharmanagar, Pecharthal, Damcherra, Kumarghat, Agartala and Udaipur) in the state of Tripura fall under the sub-tropical and temperate zones dominated by monsoons. The meteorological data was collected from the Indian Council of Agricultural Research for the period of 2012–2015 [26–28]. The fish, *C. batrachus*, were collected from different locations during April, 2012 to March, 2015. A total of 868 *C. batrachus* were examined to recover different helminth parasites. Recovered trematodes and cestodes were

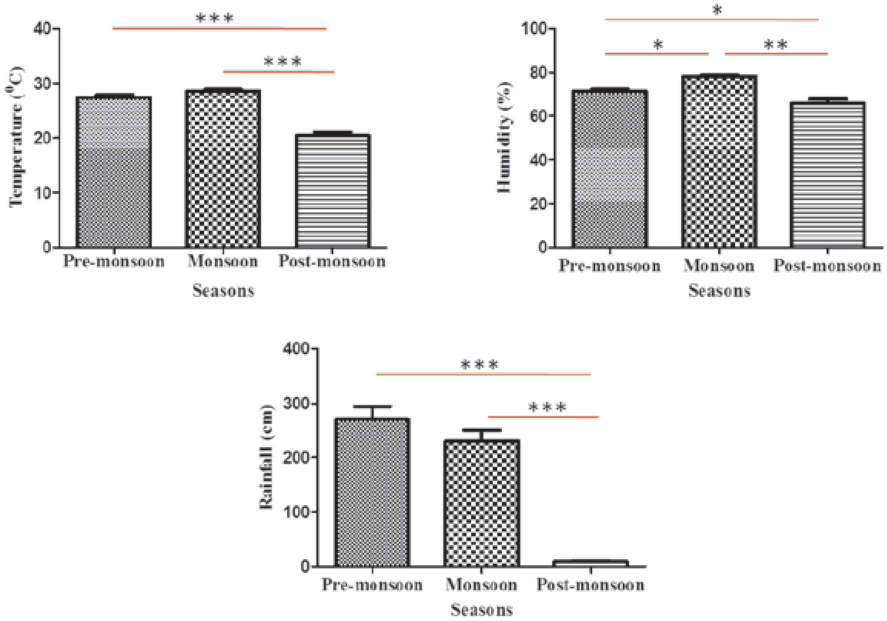

Fig. 1. Average temperature, relative humidity and rainfall of Tripura during the study period
Values are expressed as Mean ± SEM (N=12). * p≤0.05, ** p≤0.01, *** p≤0.001. One-way ANOVA, Tukey Test.

stretched over a clean slide, fixed in Neutral Buffered Formalin (NBF) and stained with borax carmine. Nematodes were preserved in 70% alcohol. They were processed through a glycerol series and mounted in glycerine jelly. The parasites were identified following Yamaguti [29–31], Jones et al. [32] and Khalil et al. [33].

The year was divided into three seasons, namely pre-monsoon (March–June), monsoon (July–October) and post-monsoon (November–February). The results were analysed for the following parameters following Margolis et al. [34]:

Prevalence (%) = number of hosts infected × 100/total number of hosts examined

Abundance = number of parasites recovered /total number of hosts examined

One-way Analysis of Variance (ANOVA) was applied to ascertain the significance of variations of the prevalence of infection between the three seasons during the study period. The ecological

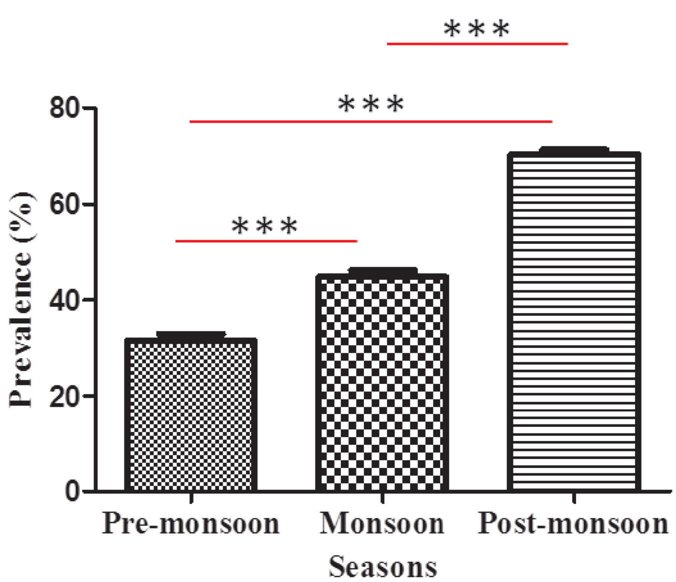

Fig. 2. Overall prevalence of helminth infections in the fish hosts examined during April, 2012 to March, 2015
Values are expressed as Mean ± SEM (N=12). *** p≤0.001. One-way ANOVA, Tukey Test.

Table 1. Prevalence, abundance and mean intensity of helminth infection in the fish hosts examined from different localities in Tripura during April, 2012 to March, 2015

Collection sites	No. of fishes examined	No of fishes infected	No. of parasites collected	Prevalence (%)	Abundance
Dharmanagar	274	206	3 083	75.18	11.25
Pecharthal	137	85	1 212	62.04	8.85
Damcherra	62	28	510	45.16	8.23
Kumarghat	70	50	272	71.43	3.89
Agartala	168	122	1 510	72.62	8.99
Udaipur	157	115	1 284	73.25	8.18
Total	868	606	7 871	69.82	9.07

relationships between the abiotic and biotic parameters (prevalence) was determined by Pearson correlation coefficients (r); p-values were calculated *vide* http://faculty.vassar.edu/lowry/tabs.html and the significance of the relationships ascertained using Bonferroni corrections.

Results

During the survey work, a total of 868 fish were collected, out of which 606 were found to be infected with various helminth parasites. A total of nine different helminth species were recovered during the study. These included one representative of the trematode group i.e., *Astiotrema reniferum*, seven representatives of the cestode group namely, *Lytocestus indicus*, *L. birmanicus*, *L. longicollis*, *L. attenuates*, *L. filiformes*, *L. clariae*, *Djombangia penetrans*, and one larval nematode species belonging to the genus *Anisakis*.

It was also observed that the overall prevalence of infection decreased significantly from the post-monsoon season to the monsoon and pre-monsoon seasons. The correlation analysis, however, revealed that the prevalence of helminth infection has a significant negative correlation with the meteorological factors like average temperature (–0.65; $p \leq 0.05$), average relative humidity (–0.70; $p \leq 0.01$) and average rainfall (–0.84; $p \leq 0.05$). In other words, the rate of infection increased with the decrease in the temperature, relative humidity and rainfall (Fig. 1–2) (Table 2).

The study showed that the fish were mainly infected with cestode parasites, followed by nematodes and trematodes (Fig. 3). The trematode *A. renniferum* was encountered only thrice during the study period, and hence this specimen was not included in the seasonal studies. It was also observed that the prevalence of cestode infection in *C. batrachus* increased from pre-monsoon to monsoon and post-monsoon season without any significant changes. However, in the case of nematodes, there was a significant ($p \leq 0.01$ and $p \leq 0.001$) increase in the infection rate from pre-

Table 2. Correlation coefficient (r) of overall prevalence of helminth infection recorded in *C. batrachus* of Tripura with meteorological parameters

	Prevalence	Temperature	Rainfall	Humidity
Prevalence	1			
Temperature	-0.65*	1		
Rainfall	-0.84***	0.96***	1	
Humidity	-0.70**	1***	0.98***	1

'-' indicates significant negative correlation (* $p \leq 0.05$, ** $p \leq 0.01$, *** $p \leq 0.001$)

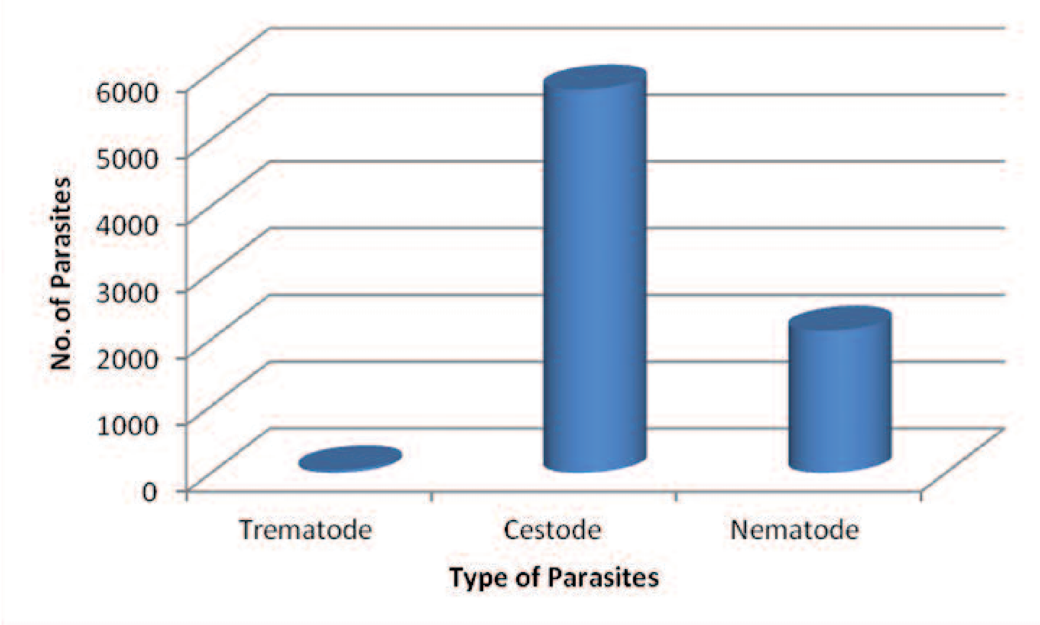

Fig. 3. Infection with different types of helminths in *C. batrachus* for the period of April, 2012 to March, 2015

monsoon to monsoon and post-monsoon season (Fig. 4). Of the seven cestodes recovered, except for *L. filiformes*, all other cestodes namely, *L. indicus*, *L. birmanicus*, *L. longicollis*, *L. attenuates*, *L. clariae* showed a significantly ($p \leq 0.05$, $p \leq 0.01$ and $p \leq 0.001$) high rate of infection in the post-monsoon season. However, in case of *D. penetrans*, the prevalence of infection was found to be highest in the pre-monsoon season with no significant difference when compared between the seasons (Fig. 5).

The prevalence of infection was also found to be highest in the fish of Dharmanagar (75.18%) followed by Udaipur (73.25%), Agartala (72.62%), Kumarghat (71.43%), Pecharthal (62.04%) and Damcherra (45.16%). The abundance value was also found to be highest in Dharmanagar (11.25) and least in Kumarghat (3.89) (Table 1).

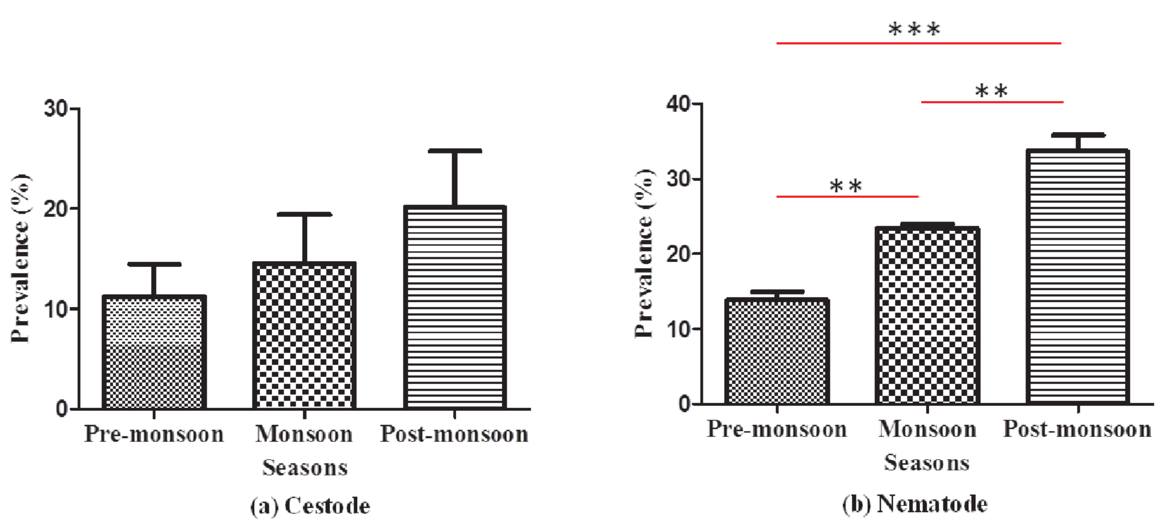

Fig. 4. Overall prevalence of (a) cestode and (b) nematode infections in the fish hosts examined during April, 2012 to March, 2015
Values are expressed as Mean ± SEM (N=12). ** $p \leq 0.01$, *** $p \leq 0.001$. One-way ANOVA,Tukey Test.

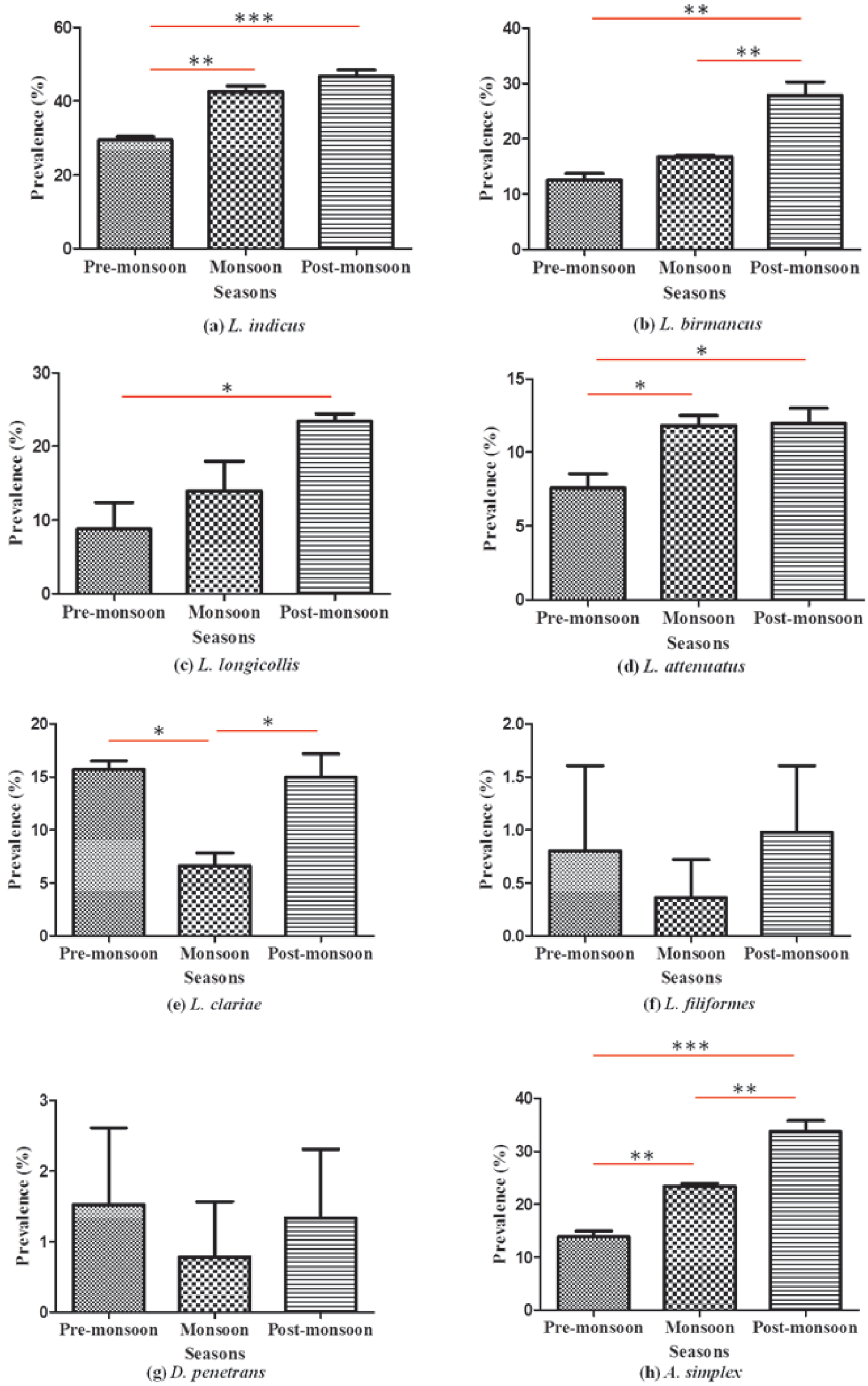

Fig. 5. Seasonal variation in the population density of (a) *L. indicus*, (b) *L. birmanicus*, (c) *L. longicollis*, (d) *L. attenuatus*, (e) *L. clariae*, (f) *L. filiformes*, (g) *D. penetrans* and (h) *Anisakis* sp. L3 larvae in *C. batrachus*
Values are expressed as Mean ± SEM (N=12). * $p \leq 0.05$, ** $p \leq 0.01$, *** $p \leq 0.001$. One-way ANOVA, Tukey Test.

Discussion

The community of helminth parasites and intensity of infection varies from species to species. Seasons interfere with the physiology and ecology of the fish, which in turn greatly influences the rate parasitic infections. The major water systems of India demonstrate a high degree of host specificity, with Siluriform fish being the most common hosts both for monozoic and segmented cestodes [35].

The results of the present study indicate that *Clarias batrachus* presented a wide diversity of helminth parasites. They also found that the percentage and intensity of helminth infection were highest in the post-monsoon season, followed by the monsoon and pre-monsoon seasons.

Among the various helminth parasites recorded from the host fish, the greatest number of infections were associated with cestodes, followed by nematodes and then trematodes. The occurrence of such a wide range of helminths indicates the presence of their intermediate and definitive hosts in the particular environment [36]. In the Himalayan region, Malhotra and Chauhan [37] observed maximum cestode infection during summer months and minimum during rainy months in various species of hill stream fishes viz. *Barilius barana*, *B. bola*, *B. vagra*, *B. bendelisis*, *Garra gotyla gotyla*, *Glyptothorax telchitta*, *Heteropneustes fossilis*, *Labeo calbasu*, *L. dyocheilus*, *L. rohita*, *Schizothorax plagiostomus* and *S. richardsonii*. Several authors have reported a number of Caryophyllidean species belonging to the genus *Lyocestus* and *Djombangia* infecting the walking catfishes of Northeast India and its adjacent region [18–21,38]. Among the cestode parasites recorded in the present study, *L. indicus*, *L. birmanicus*, *L. longicollis* and *L. clariae* occurred more frequently than *L. attenuatus*, *L. filiformes* and *D. penetrans.* A similar prevalence was also observed by Chakravarty and Tandon [18] in the case of infection with *L. indicus.* The results of the present study differ from the observations made by Chakravarty and Tandon [18], which found *L. birmanicus* and *L. longicollis* to be dominant, while *D. penetrans* occurred concurrently with the dominant species.

In the present study, *C. batrachus* was found to be infected with nematodes in all seasons but with a peak in the post-monsoon season, suggesting a seasonal fluctuation in infection. Continuous recruitment and development of the larval Anisakid nematodes may occur in *C. batrachus*, although parasite reproduction may be at its peak in the monsoon season. This result agrees with the observations made on nematodes infecting *Mugil cephalus* in Saloum and Senegal rivers [39]. Skorping [40] explained that non-seasonal patterns in infection levels result from an overlap in the seasonal rates of nematode mortality and recruitment, and concluded that small shifts in these rates would lead to more pronounced seasonal patterns. This hypothesis may explain differences in the seasonal changes of infection as observed in the present study due to nematodes in different localities.

Seasonal variations in populations of fish parasites have been observed by several authors, including Kennedy [41], Furtado and Tan [42] and Moravec [43], and summarized by Chubb [44], that seasonal variation in water temperature is one of the dominant factors acting directly or indirectly on the parasite, by affecting host behaviour and metabolism. In the present study, the rate of infection increased with decrease in temperature, and a temperature range of 19–20°C was found to be favourable for the propagation of fish parasites in the study area. A similar observation was made by Kundu et al. [45] in the *Channa punctatus* infected with *P. allahabadii* and concluded that the immune response drops with the decrease in temperature, making the fish more vulnerable to parasites. Parasitic infection increases with organic enrichment of the water bodies caused by pollution, agricultural runoff and excessive use of antibiotics, which increases the density of intermediate hosts. Therefore, the combination of low metabolic activity and the suppression of the natural immune system of the host fish increases their susceptibility to a wide range of parasitic diseases [46]. Changes in the prevalence and mean intensity of parasitic infection might be influenced by various factors such as water pollution, parasite biology, host hormonal status, host immunological response, host migration, changes in the feeding habits of the host and the availability of infected intermediate hosts [47–50]. However, the influence of these factors is difficult to distinguish because they most likely interrelated and influence each other.

Conclusions

The findings of the study indicate that climate changes in aquatic systems affect most organisms and their functional roles in the ecosystem. Changes in these roles may be difficult to detect, but our examination of parasite communities in fish has afforded an insight into any structural and functional alterations in the system. Our findings indicate that climatic conditions such as temperature, relative humidity and rainfall play a major role in determining the density of parasitic infection by interfering with the life cycles of the parasite and their intermediate hosts, as well as the metabolism of the final host.

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