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Gyrodactylus in the spotlight: how exposure to light impacts disease and the feeding behavior of the freshwater tropical guppy (*Poecilia reticulata*)

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Abstract

Artificial light at night (ALAN) negatively impacts organisms in many ways, from their feeding behaviors to their response and ability to deal with disease. Our knowledge of ALAN is focused on hosts, but we must also consider their parasites, which constitute half of all described animal species. Here, we assessed the impact of light exposure on a model host–parasite system (*Poecilia reticulata* and the ectoparasitic monogenean *Gyrodactylus turnbulli*). First, parasite-free fish were exposed to 12:12 h light:dark (control) or 24:0 h light:dark (ALAN) for 21 days followed by experimental infection. Second, naturally acquired *G. turnbulli* infections were monitored for 28 days during exposure of their hosts to a specified light regime (6:18 h, 12:12 h, or 24:0 h light:dark). Experimentally infected fish exposed to constant light had, on average, a greater maximum parasite burden than controls, but no other measured parasite metrics were impacted. Host feeding behavior was also significantly affected: fish under ALAN fed faster and took more bites than controls, whilst fish exposed to reduced light fed slower. Thus, ALAN can impact parasite burdens, even in the short term, and altering light conditions will impact fish feeding behavior. Such responses could initiate disease outbreaks or perturb food-webs with wider ecological impacts.

KEYWORDS
ALAN, Feeding Behaviour, Freshwater Diseas, *Gyrodactylus*

1 | INTRODUCTION

Urbanization has led to prolonged exposure of organisms to artificial light, particularly artificial light at night (ALAN) (Cesarz et al., 2023; Falcón et al., 2020). This light may either be spillover from urban areas, still allowing organisms to experience some level of diurnal light change, or more controlled light manipulation, as might be the case in aquaculture (Wang et al., 2023). Both have the potential to disrupt or alter various biological functions, including flight patterns, mating and orientation, and maturation, all of which are dictated by light cues (Aulsebrook et al., 2018; Dechaine et al., 2009; Muheim et al., 2006; Sweeney et al., 2003). Despite being a known issue, the drivers behind changes in ALAN often revolve around socio-economic issues, such as energy costs and greenhouse gases, rather than addressing the necessity to reduce environmental stressors on ecosystems (Holker et al., 2021; Hooker et al., 2022; Stone et al., 2012). Light intensity, quality, color, and duration all impact organisms and their interactions with the environment (Boeuf & Le

Bail, 1999). This complexity is amplified in variable aquatic environments, where organisms constantly move within three dimensions and light is reflected, refracted, and attenuated as depth and turbidity increase (Evans, 2004; Stoner, 2004; Sumpter, 1992). Photoreception by fish occurs via both the eyes and the pineal gland, and is species and life-stage dependent (Tabata, 1992). Most species require minimal light exposure per day to develop and grow, linked to activities like movement, hunting, and visual foraging (e.g. Iigo & Tabata, 1997; Richardson & McCleave, 1974). In aquaculture, light exposure is intentionally increased to boost production (Wang et al., 2023). Overexposure to light, however, can increase stress in fish (Ellison et al., 2021; Tian et al., 2015) and even result in mortalities (Schligler et al., 2021). ALAN has been shown to reduce sex steroids and mRNA expression of gonadotropins in European roach (*Rutilus rutilus* L.) and perch (*Perca fluviatilis* L.) (see Bruning et al., 2018). Behaviourally, ALAN can increase fish predation on invertebrate prey and alter the emergence of fish (Czarnecka et al., 2019; Kurvers et al., 2018).

All fish are infected by a range of naturally occurring parasites, which often only become detrimental to the host following environmental perturbation (Wood & Johnson, 2015). We know ALAN can influence interactions between species, such as foraging and predation, but less is known about the impacts of ALAN on parasites (Dwyer et al., 2013; Rodríguez et al., 2017; Rodríguez et al., 2021; Rydell, 1992). While exposure to increased photoperiods can indirectly exacerbate the problem of infectious diseases (e.g. via reduced immunity in fish [Bakke et al., 2021; Ellison et al., 2021; Tian et al., 2015] or increased parasite reproduction [Gannicott & Tinsley, 1997]), parasites are also directly impacted by light and exhibit daily rhythmicity in gene expression (de Bekker et al., 2017; Hunt et al., 2022; Rijo-Ferreira et al., 2020; Smith et al., 2020). For macroparasites, such as *Argulus* spp. (aquatic lice), we know that they display diurnal behavioral patterns off-host, with males showing increased activity at the change of light, while females exhibit a delayed pattern (Hunt et al., 2021). The microparasite *Gyrodactylus turnbulli* (an ectoparasitic monogenean) is more active in the dark and it even induces restlessness in its host, the guppy *Poecilia reticulata* (Peters 1859) (hereafter 'guppy'), particularly at night (Arapi et al., under review). It is with this non-invasive freshwater model that we continue to investigate how light can impact the relationship between parasites and their hosts.

From an aquaculture perspective, we posed the following two questions. Do altered light regimes impact infection dynamics between the guppy and *G. turnbulli*? Is the feeding behavior of guppies (both infected and uninfected) impacted by different light regime exposure? These questions were investigated under laboratory conditions: firstly, experimental infection with *G. turnbulli* was assessed under control light (12:12 h light:dark) and total light regime and, secondly, naturally acquired infections of *G. turnbulli*, along with host feeding behavior, were assessed under three different light regimes (6:18 h, 12:12 h, and 24:0 h light:dark). In both instances, exposure to light alterations was relatively short term (3–5.5 weeks).

2 | METHODS

2.1 | Host–parasite system

We utilized the established guppy–*Gyrodactylus* system for this study. The guppy (*Poecilia reticulata*) is a well-studied model fish host and the genus *Gyrodactylus* is a species-rich group of fish pathogens that are both ecologically and economically important and globally pervasive (Bakke et al., 2007). *G. turnbulli* is the primary monogenean ectoparasite of guppies and a major pest in the ornamental fish trade, known as a "Russian-Doll parasite" due to its hyperviviparous reproductive capabilities (Bakke et al., 2007). One key benefit of this parasite–host system as an experimental system is the ability to non-invasively track individual host parasite burdens over time.

Size-matched (254–294 mm) mixed ornamental adult female guppies were imported to Cardiff University ($n = 300$) and acclimated in shoals of 10–15 fish for 24 h, maintained within 40-L aquaria at 24 ± 0.5°C on a 12:12 h light:dark photoperiod (500 lux, lights abruptly on 7 a.m. and off at 7 p.m.). This stock was used for both experiments. All fish were screened to assess ectoparasite communities. All fish were confirmed to host *G. turnbulli* through screening (position of worms was an indicator of species, then samples were sequenced via PCR for confirmation following the protocol outlined in Schelkle et al. [2012]). Briefly this involved extraction of DNA using a Qiagen DNeasy blood & tissue kit following the manufacturer's instructions, using primers from Harris et al. (1999) which amplified the 5.8 s gene and partial ITS-1 and ITS-2 regions (R1 = ACTCCATG- TGGTGGATC and F3 = TTGCTGCACTCTTCATC). The PCR of extracted DNA was performed as described by Faria et al. (2011), and extracted DNA was sent to Eurofins Genomics for sequencing (10 μL of nuclease-free water, 5 μL of PCR product, and 2 μL of each primer at 10 μM concentration). The resulting contigs for each sample were then subject to an NCBI BLAST search for related sequences, with confirmation of species taken on a match of 100% sequence identity to *G. turnbulli*.

Prior to the investigation, $n = 210$ fish were treated with Levamisole (Norbrook®) according to Schelkle et al. (2009) and $n = 90$ retained with their parasite load for the infected treatments in Experiment 2. The treated fish were then screened and confirmed free of ectoparasites if no parasites were detected through microscopic examination three consecutive times (see Schelkle et al., 2009). This involved mildly anesthetizing individual fish using 0.02% MS-222 and observing the surface of each fish for visible signs of parasitaemia (e.g., raised fins, white spots, abnormal growths), where any infected fish were excluded. The remaining fish ($n = 90$) were monitored daily for signs of abnormalities or mortalities. All fish, prior to experimental infections, were measured for standard length by mildly anesthetising individuals as above. All fish were then individually isolated into 1-L aquaria (for the duration of both experiments) and maintained under the same environmental conditions as before (24 ± 1°C on a 12:12 h light:dark photoperiod) for 3 days until the beginning of the experiment. Fish were fed twice daily on flake food (Aquarian®) 1 day and freshly hatched *Artemia* nauplii on alternate days. Feeding regimes were maintained throughout, and any/all mortalities of either infected

or uninfected fish were recorded. As the infection trajectory for individual fish was followed, pseudo-replication was accounted for during analysis (see the Statistical analysis section below).

2.2 | Experiment 1: Impact of ALAN on experimental *G. turnbulli* infections

Female ornamental guppies ($n = 120$, all confirmed parasite free) were separated into two groups ($n = 60$) and exposed to either 12:12 h light:dark (control light of 500 lux) or 24 h light (artificial light at night, ALAN) for 21 days. Lighting consisted of Sylvania standard fluorescent bulbs, the intensity of which were measured using a Testo 540 Pocket Light Meter at the centre of the 1-L aquaria ($n = 10$ replicates per group). The lighting intensity range matched that of an aquaculture optimum (~500 lux; Qu et al., 2022). Then, half of the fish ($n = 30$) from each regime were infected with *G. turnbulli* according to Schelkle et al. (2009). Briefly, this involved anesthetizing recipient fish lightly with 0.02% MS-222 and bringing the recipient gently towards a deceased donor until two individual *G. turnbulli* attached onto the recipient's caudal fin. Every two sequential days following initial exposure all infected fish had their parasite numbers assessed through mildly anesthetising infected fish (using 0.02% MS-222) and counting the number of worms present under a dissecting microscope with fiberoptic illumination until day 19, when most fish had cleared their infection or host mortality had occurred. The uninfected fish were handled in exactly the same manner as the infected fish throughout the experiment, starting with sham infections (anesthetised and handled but without the introduction of parasites).

2.3 | Experiment 2a: Impact of differing light regimes on established *G. turnbulli* infections

Fish ($n = 60$ per regime, $n = 180$ total) were exposed to one of three light (500 lux) regimes: 6:18 h (short day), 12:12 h (control), or 24:0 h (long day/ALAN). Within each regime, $n = 30$ fish were host to variable naturally acquired *G. turnbulli* infections (all were pre-quantified, range 8–153 worms per host) and $n = 30$ were confirmed ectoparasite clear to act as uninfected controls to account for exposure mortality. Every two sequential days following initial light exposure, all infected fish had their parasite numbers assessed as in Experiment 1. The last parasite screen occurred on day 28, when almost all fish had cleared their infections or died.

2.4 | Experiment 2b: Impact of differing light regimes on host feeding behavior

To understand the effect of light exposure on the feeding behavior of fish, feed trials were conducted once a week during the experiment for both infected and uninfected fish. Fish were fed as in Experiment 1. Feeding trials involved introducing 1 mL of *A. salina* nauplii into the individual fish aquaria, illuminated under artificial light (500 lux, identical to pre-exposure lighting) equally to minimize shadowing, commencing 3 h after lights on. On immediate introduction, feeding latency (time from the introduction of food until the first bite) was noted, after which the feeding rate (number of feed lunges occurring within 30 s after the first bite) was recorded. Unsuccessful lunges towards individuals of *A. salina* were still recorded as bites, and occurrences where the *A. salina* were eaten, expelled, and re-ingested were counted as separate bites. Following the feeding trials, all fish were returned to their experimental light conditions.

2.5 | Ethics statement

All animal work was approved by the Cardiff University Animal Ethics Committee and conducted under UK Home Office license PP8167141.

2.6 | Statistical analysis

All statistical analyses were carried out using Rstudio version 4.2.3. (http://www.R-project.org/) and the following packages: 'MASS' for general and generalized linear models and data transformation, 'ggplot2' for visualization of data, 'lme4' for general and generalized linear mixed models and 'effects' for graphical overviews of data.

For analysis of infection data (for both Experiments 1 and 2) the following variables were measured in regard to parasite metrics: the area under the curve (AUC), duration of infection, host death day, maximum parasite burden, peak day, death day, and mean parasite intensity. To calculate AUC, a common parasite metric which quantifies overall pathogen burdens across a known trajectory, we utilized the trapezoid rule (White, 2011). Duration of infection was classified as the time until a fish either cleared its parasite burden or died (host death day). The maximum parasite burden was defined as the highest number of *G. turnbulli* a single host achieved on a single day during the duration of the experiment, the day of which was defined as the peak day. Mean parasite intensity was taken as the average number of *G. turnbulli* worms remaining within the system across the duration of the experiment. Standard length was initially included in all statistical models, but as it did not explain significant variation it was removed from subsequent models as part of model refinement (Thomas et al., 2013). For analysis of the summation of all AUC values (AUC.sum), we transformed the data using the Box–Cox transformation method within the *MASS* package in R, as no combination of family or link functions could satisfy the assumptions of the general linear model (GLM) prior to transformation. Following transformation, a GLM using the Gaussian family and identity link functions was run to investigate how AUC varied with regime. To analyze how maximum parasite count varied between light regimes, we used a generalized linear model (GisedLM) with a negative binomial family and identity link function. For the analysis of variation between peak day and light regime, a Box–Cox transformation was again used, and a GisedLM using the inverse Gaussian family and identity link function run. Infection duration variation between regimes was analyzed with a GLM using a Gaussian family and identity link function. For the analysis of

mean parasite intensity and account for pseudo replication in regards to individual fish, a generalized linear mixed model (GisedLMM) in the 'lme4' package was used with a negative binomial family and identity link function. Within this model, parasite number was our response variable, and regime and the interaction between regime and time our dependent variables and individual fish ID was our random factor.

For the analysis of the host feeding ecology data (Experiment 2), latency to feed was analyzed against regime and the interaction between week and regime using a GisedLMM. For this, latency was our response variable, with light regime, week, and the interaction between the two as our dependant variables and fish ID as our random variable, with the Gaussian family and sqrt link function. For analysis of the number of bites over time, we used a GisedLMM with a Poisson family and a log-link function link. The analysis of both maximum bite count and mean bite count per fish was carried out using a GisedLM with the negative binomial family and identity link function.

3 | RESULTS

In both experiments for all light regimes, parasite burden on the individually isolated hosts varied significantly with time (GisedLMM: $df = 3$, $\chi^2 = 1618.49$, $p < 0.0001$). Time was also a significant factor for all treatments with regard to feeding latency (GLMM: SE = 0.02, $z = -34.75$, $p < 0.0001$).

3.1 | Experiment 1: Impact of ALAN on experimental *G. turnbulli* infections

Fish exposed to both ALAN (24:0 h light:dark) and controls (12:12 h light:dark) exhibited similar parasite burden trends, increasing until a peak and then decreasing (Figure 1a), but the maximum parasite count varied significantly, with fish exposed to 24 h of light (ALAN) having a greater maximum parasite burden than those under 12 h of light (GisedLM: SE = 16.87, $t = 2.20$, $p = 0.02$). For mean parasite intensity across the experiment, fish exposed to 24 h of light did not significantly vary compared to controls (GisedLMM: SE = 0.93, $z = 0.018$, $p = 0.98$). Similarly, the AUC.sum, duration of infection, peak day or death day did not significantly vary between treatments (see Table 1 for statistical outputs).

3.2 | Experiment 2: Impact of differing light regimes on established *G. turnbulli* infections

At the start of the experiment, the mean number of gyrodactylids per treatment group was equal ($n = 65$), with individual host burdens

TABLE 1 Statistical outputs for parasite parameters (AUC, maximum parasite count, peak day, infection duration, parasite abundance, death day, and parasite count) for the 24-h treatment in Experiment 1, using control as a baseline.

Parameter tested	Standard error	*t*/*z* value	*p* value
AUC.sum	0.0065	0.17	0.86
Maximum parasite burden	16.87	2.20	0.02*
Peak day	1.35	1.36	0.18
Infection duration	0.04	−0.59	0.5
Parasite abundance	3.90	1.30	0.19
Death day	4.86	1.61	0.15
Parasite count	0.93	0.01	0.98

Note: Included are the standard errors, the *t* or *z* value (test dependent) and *p* value for all parameters tested. *denotes significance.
Abbreviations: AUC, area under the curve; AUC.sum, summation of all AUC values.

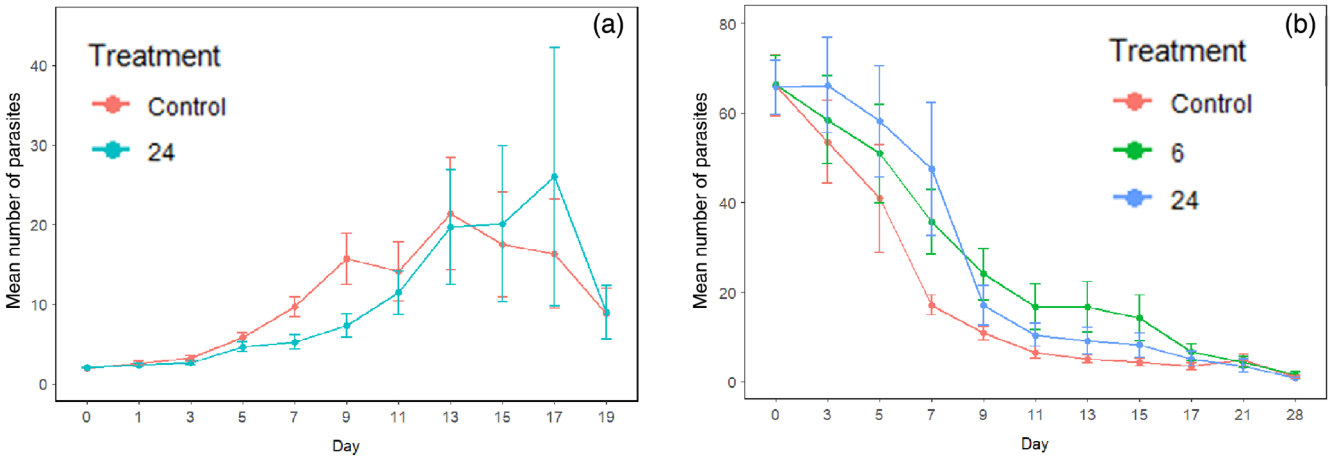

FIGURE 1 Mean parasite intensities of *Gyrodactylus turnbulli* per light regime (distinguished by color) per day (including standard error) on their host *Poecilia reticulata*. (a) Experiment 1, where fish were exposed to either control (12:12 h light:dark) or ALAN (24:0 h light:dark) light regimes over 19 days. All naïve fish in the two treatments were experimentally infected with two individual parasites on day 0 after 21 days pre-exposure to the different light conditions. (b) Experiment 2, where fish were exposed to control (12:12 h light:dark), low light (6:18 h light:dark), or ALAN (24:0 h light:dark) light regimes over 28 days starting with naturally acquired infections with a mean starting infection level in the three different treatments of 65 worms per treatment. In this experiment, fish were all exposed to the same control light conditions until day 0.

ranging from eight to 153 worms (Figure 1b, day 0). As expected from naturally acquired infections, when the hosts were isolated the parasite burden generally declined with time (there were exceptions where certain individuals were particularly susceptible [$n = 15$ total, $n = 5$ per treatment] and their burden increased until mortality occurred). When compared to the control treatment (12 h of light), none of the measured parasite metrics (AUC, maximum parasite count, average parasite burden, peak day, and duration of infection) varied significantly between treatments (Table 2). When looking at parasite count over time (accounting for pseudoreplication), there was no significant difference between treatments (GLMM: 6 h; SE = 0.25, $z = 1.08$, $p = 0.28$, 24 h; SE = 0.25, $z = 1.63$, $p = 0.10$).

3.3 | Experiment 2: Fish feeding behavior

Latency to feed was impacted by altered light exposure (Table 3), with fish exposed to 24 h of light responding significantly faster (shorter latency) than the controls (GLMM: SE = 0.14, $z = -10.79$, $p < 0.0001$), whilst those exposed to 6 h of light fed significantly more slowly (longer latency) compared to controls (12 h) (GLMM: SE = 0.14, $z = 2.98$, $p = 0.002$). Fish exposed to 24 h of light took significantly more bites than controls (GLMM: SE = 0.07, $z = 8.30$, $p < 0.0001$) and decreased their bite count over time (GLMM: SE = 0.017, $z = -13.36$, $p < 0.0001$), whereas the bite count of fish exposed to 6 h of light remained constant (GLMM: SE = 0.019, $z = -0.39$, $p = 0.70$) (Figure 2). Neither the maximum bite count nor the mean bite count per fish significantly varied between treatments (GisedLM maximum bite count: 6; SE = 2.96, $t = -1.58$, $p = 0.12$, 24; SE = 3.13, $t = 1.18$, $p = 0.24$, GisedLM mean bite count: 6; SE = 2.44, $t = -1.26$, $p = 0.21$, 24; SE = 2.57, $t = 0.90$, $p = 0.37$). Infection with *G. turnbulli* did not impact feeding behaviors (GisedLMM latency to feed: SE = 0.11, $z = -0.76$, $p = 0.44$, GisedLMM number of bites: SE = 0.05, $z = 0.73$, $p = 0.48$).

4 | DISCUSSION

We sought to assess the relationship between fish exposure to ALAN (24:0 h light:dark) and infection with the ectoparasitic monogenean *G. turnbulli*. Exposure to ALAN following experimental infection resulted in increased maximum parasite burdens but did not significantly impact any other measured parasite metric. Fish feeding behavior was also influenced, with fish exposed to ALAN showing a shorter latency to feed and increased bite count, whilst those exposed to shorter light periods (6:18 h light:dark) had a longer latency to feed.

Chronic light exposure has been shown previously to significantly impact gyrodactylid dynamics in controlled laboratory settings. Subjecting laboratory-reared three-spined sticklebacks (*Gasterosteus aculeatus* L.) to altered light conditions (specifically, 6 months under a 16:8 h light:dark cycle) resulted in increased susceptibility to *Gyrodactylus gasterostei* (see Whiting et al. [2020]). More acutely, we found experimentally infected guppies exposed to ALAN for 21 days prior to infection experienced a significantly higher maximum parasite burden. All other parasite metrics analyzed here showed no significant changes in response to altered photoperiod. Manipulating the photoperiod, however, even in the short term, can influence fish immunity and induce stress (Bowden, 2008; Ellison et al., 2021), factors that likely contribute to the observed increase in maximum parasite burden. Direct comparisons with other host–parasite systems, however, are problematic, especially when comparing temperate (*G. gasterostei*) and tropical (*G. turnbulli*) species. The temperature-dependent nature

TABLE 2 Statistical outputs for parasite parameters (AUC, maximum parasite count, peak day, infection duration, average worm count, and parasite count) for Experiment 2, using control as a baseline.

Parameter tested	Treatment	Standard error	*t*/*z* value	*p* value
AUC.sum	6	0.07098	1.087	0.280
	24	0.07098	1.424	0.158
Maximum parasite burden	6	13.954	0.148	0.882
	24	14.865	0.868	0.385
Peak day	6	0.7944	1.762	0.0815
	24	0.6635	0.955	0.3424
Infection duration	6	0.14433	0.418	0.677
	24	0.14433	0.742	0.460
Average parasite count	6	5.806	0.810	0.418
	24	6.031	1.117	0.264
Parasite count	Treatment	Df 2	ChiSq 2.75	0.25
	Treatment: day	Df 3	ChiSq 1618.49	<0.0001*
	6	0.25	1.08	0.28
	24	0.25	1.63	0.10

Note: Included are the standard errors, the *t* or *z* value (test dependent) and *p* value for all parameters tested. *denotes significance.
Abbreviations: AUC, area under the curve; AUC.sum, summation of all AUC values; Df, degrees of freedom; ChiSq, chi squared value.

TABLE 3 Statistical outputs for feeding behaviors (latency to feed, number of bites in 30 s, maximum bite count, and mean bite count) for Experiment 2, using control as a baseline.

Parameter tested	Treatment	Standard error	*t*/*z* value	*p* value
Latency to feed	6	0.14	2.98	0.002*
	24	0.14	−10.79	<0.0001*
	6:week	0.03	−4.09	<0.0001*
	24:week	0.02	28.07	<0.0001*
	Infection	0.11	−0.76	0.44
	Week	0.02	−34.75	<0.0001*
Number of bites	6	0.08	−1.18	0.23
	24	0.07	8.30	<0.0001*
	6:week	0.019	−0.39	0.70
	24:week	0.017	−13.68	<0.0001*
	Infection	0.05	0.73	0.48
	Week	0.01	14.52	<0.0001*
Maximum bite count	6	2.96	−1.58	0.12
	24	3.13	1.18	0.24
Average bite count	6	2.44	−1.26	0.21
	24	2.57	0.90	0.37

Note: Included are the standard errors, the *t* or *z* value (test dependent) and *p* value for all parameters tested. *denotes significance.
Abbreviations: 6:Week is the interaction between the 6 hour light regime and the week feeding was assessed; 24:Week is the interaction between the 24 hour light regime and the week feedng was assessed.

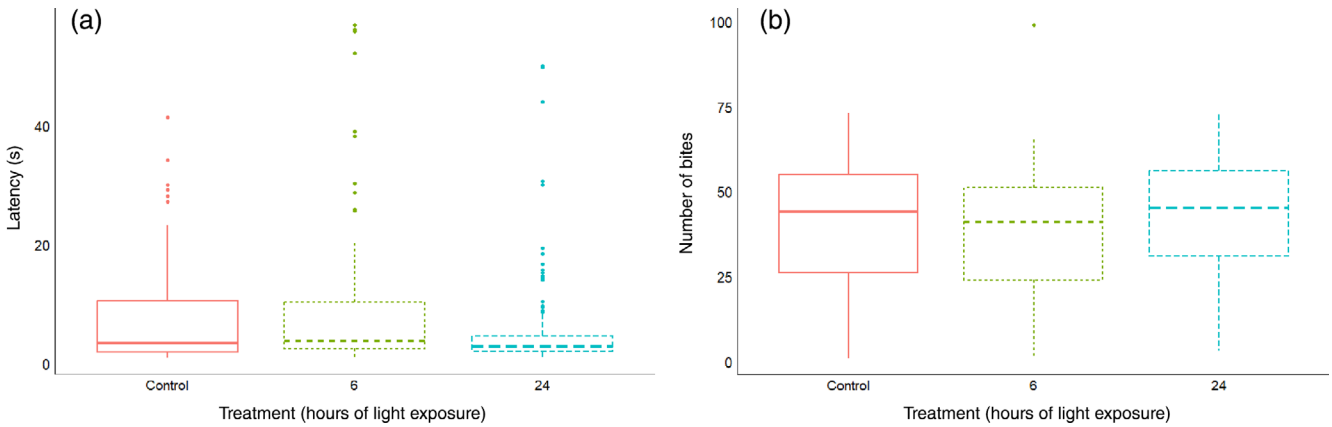

FIGURE 2 Feeding behavior of *Poecilia reticulata* in Experiment 2. (a) Latency (in seconds) to first bite per treatment across the duration of the experiment. (b) Bite count (recorded as number of bites taken within 30 s) per treatment across the duration of the experiment. Box plots show the median (line), interquartile range (box), and the 1.5× interquartile range (whiskers). Any filled circles represent values outside the 1.5× interquartile range.

of parasite reproduction (and host immunity) affects the timing of their response to any environmental perturbation. In Experiment 2, we observed a steady decrease in parasite numbers over time, reflecting the parasites' inability to sustain infrapopulations on immunologically active fish. This decline was not affected by increased light, presumably due to the overriding impact of host immunity. Such parasite trajectories are representative of infections that have past their peak, in contrast to newly established experimental laboratory infections (e.g., King & Cable, 2007; Masud et al., 2020; Smallbone et al., 2016; Stewart et al., 2017). We also know that the activity of gyrodactylids increases in the dark, as demonstrated by *G. gastorostei* off the host (Brooker et al., 2011) and by *G. turnbulli* on the host (Arapi et al., under review). In the latter case, this heightened activity results in restlessness of their diurnal hosts (Arapi et al., under review), another factor which might have been at play in the current experiments.

Light is vital to fish that are visual hunters (López-Olmeda & Sánchez-Vázquez, 2010). Diurnal feeders rely on light to illuminate prey, which are then more vulnerable to predation. The impact of ALAN could worsen this, and heightened light can also lead to increased

invertebrate emergence, potentially disturbing food webs (Czarnecka et al., 2019; Kurvers et al., 2018). We found both fish latency to feed and bite count were impacted by altering light regimes despite all our feeding behavior recordings being conducted under well-lit conditions, consistent between regimes. As there was also no enrichment in the tanks to obstruct the prey, the vision (Diehl, 1988; Aksnes & Giske, 1993; Brooker et al., 2011) and olfaction of the fish are unlikely to have been impacted (Stoner, 2004). Our ALAN-exposed fish showed the shortest feeding latency, consistent with prior research on light impacting fish activity (Marchand et al., 2003; Scherer & Harrison, 1988; Stoner, 2004; Trippel & Neil, 2003). Increased light exposure also correlates with heightened fish activity and feeding (Czarnecka et al., 2019) as more energy is required to maintain this level of activity (Adegboye et al., 2017, 2020). Increased activity may reduce overall fitness and increase susceptibility to parasites if the increased nutrient intake does not sufficiently compensate for the expended energy. As *G. turnbulli* interferes with host resting (Arapi et al., 2024), this potentially explains the lack of significance in parasite metrics between infected fish under 6:18 h light:dark and those under ALAN. The observed latency in fish exposed to 6 h of light and 18 h of darkness could be due to lethargy, resulting in reduced activity (also shown in Jones et al., 2017; Jones & Hale, 2020). During the current study, we found no impact of short-term light exposure on host mortality. In contrast, long-term exposure (>18 months) to ALAN (4.3 lux) decreased average survival in anemonefish (*Amphirion chrysopterus* Cuvier 1816) by 36%, and decreased their growth (Schligler et al., 2021).

In aquaculture, light is manipulated for economic benefit (Frenzl et al., 2014; Hou et al., 2019), but hobbyists and pet owners are often encouraged to illuminate their aquarium tanks 24 h a day. Informed adjustment of light regimes could potentially reduce parasite infection by optimizing photoperiods to enhance fish growth while minimizing or negatively impacting parasite infections. The potential for light as a disease management tool (chronotherapy) would heavily depend on the host–parasite system. In terrestrial environments, increased red light exposure for plants increased their salicylic acid levels, a defense mechanism against infection (Gallé et al., 2021). Oomycete pathogens sporulate in darkness, and therefore the artificial lighting used in aquaculture may inadvertently inhibit their sporulation, growth, and/or propagation (Cohen et al., 1975; MacAulay et al., unpublished; Xiang & Judeslon, 2014). Chronotherapy could also enhance treatment efficacy, for instance by guiding the timing of treatments for *Argulus foliaceous* based on when they are most vulnerable. This approach is supported by the discovery that the genes targeted by treatments display a daily rhythmicity (Hunt et al., 2022). Manipulation of photoperiod offers a non-invasive mitigation strategy for aquaculture to improve pathogen mitigation and provides a counterargument to the drawbacks of extended photoperiods on fish health; unfortunately, this is likely very host–pathogen specific.

Our work adds to the data on ALAN impacting fish, in particular increased host feeding activity prior to and during infection with *G. turnbulli*. Here, despite the relatively short duration of exposure we observed mild effects of acute ALAN exposure on maximum parasite burdens of a freshwater fish host, whilst all other parasite metrics were not significantly impacted. Together with the impact on host feeding, this study highlights the implications chronic light exposure may have on host–parasite dynamics, especially with regard to multiple stressors.

AUTHOR CONTRIBUTIONS

Scott MacAulay conceived and carried out all work and analysis. Jo Cable supervised and reviewed the manuscript.

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DATA AVAILABILITY STATEMENT

Data is available at https://doi.org/10.17035/d.2024.0321119798.

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