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Research Article

Effects of a prophylactic, intermittent, long-term formalin treatment on channel catfish, *Ictalurus punctatus* (Actinopterygii: Siluriformes: Ictaluridae) growth

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ABSTRACT. Prophylactic chemical treatments are commonly used in fish farms to prevent and control infectious diseases, with formalin being one such chemical. The purpose of this study was to see how an eight-week trial of intermittent, prophylactic, low-dose (25 mg L^{-1}), 1-h, and biweekly formalin baths affected channel catfish (*Ictalurus punctatus*) growth indices (mean weight, condition index, specific growth rate (SGR), feed conversion ratio, and feed consumption). Most growth indices were not statistically significant between the treated and control fish at the end of the trial, except for total SGR, where formalin-treated fish had a significantly higher SGR ($P < 0.05$) and steeper growth curve compared to control fish. In conclusion, intermittent and long-term use of formalin at low doses has no adverse effect on channel catfish growth.

Keywords: *Ictalurus punctatus*; channel catfish; formalin; prophylactic; long-term; treatment

INTRODUCTION

Chemicals used to control infectious diseases in aquaculture, as well as their doses and delivery methods, vary depending on the pathogen and water quality, as their efficacy can be influenced by environmental factors and the specific fish species (Bondad-Reantaso et al. 2023). Due to its low cost and high effectiveness, formaldehyde is a widely used chemical (de la Gándara et al. 2002). It is typically marketed for aquaculture as formalin, a 37% aqueous solution of formaldehyde (Wiegers et al. 2006). Formalin is approved in the USA (FDA 2024) for the control of external parasites and fungi in cultured fish (Rowland et al. 2006, 2008, LaPatra & MacMillan 2008, Jin et al. 2010, Pahor-Filho et al. 2015), and it may also be used to increase egg survival during hatching (Rasowo et al. 2007).

Formalin therapeutic doses can range from 1,000 to 2,000 mg L^{-1} for 15 min to control egg fungal infections, 12.5-25 mg L^{-1} in long-term bath treatments, or 250 mg L^{-1} in short-term baths of up to 30-60 min (Wiegers et al. 2006). In the long term, intermittent prophylactic chemical administration to cultured fish may have negative growth consequences (Powell et al. 1994). However, the intermittent use of formalin in cold-water fish at therapeutic doses does not appear to affect the growth of salmonids (Jimoh et al. 2020), even when used repeatedly in Arctic charr (*Salvelinus alpinus*) (Kristjansson et al. 1995). According to a recent article on the use of formalin in intensive aqua-

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culture, its main effect is on fish tissues (Leal et al. 2018). Furthermore, epithelial cell damage (Buchmann et al. 2004), possible immunosuppressive effects (Holladay et al. 2010), as well as genotoxic and cytotoxic effects in fish have been documented after formalin immersion (Jerbi et al. 2011).

Several compounds and antibiotics are used to improve and prevent disease emergence, but there are limitations to the use of antibiotics and chemicals in aquaculture species (Costello et al. 2001). Furthermore, there have been few studies on the effects of long-term, intermittent, and low therapeutic doses of formalin on warm-water aquaculture, including channel catfish (*Ictalurus punctatus*), since formalin is more toxic to channel catfish at higher temperatures (Piper 1982). As a result, the goal of this study was to see how prophylactic, intermittent, and long-term use of formalin affected the growth rates of *I. punctatus* under controlled conditions.

MATERIALS AND METHODS

A stock of 180 channel catfish fingerlings (4-6 cm furcal length) obtained from a fish hatchery in Tamaulipas (Mexico) and transported to the laboratory in the Facultad de Medicina Veterinaria y Zootecnia (Universidad Autónoma de Tamaulipas), were placed in six round 50 L plastic tanks filled with fresh water to a volume of 46 L. Each tank received aeration from a blower (Sweetwater®) and a constant water supply (3 mL s^{-1}). A biofilter, consisting of a nylon screen, polyester fiber, and activated carbon, was attached to each tank. During the trial, water flow increased (up to 6 mL s^{-1}) as the fish biomass in the tank increased. Weekly water parameters were measured using a freshwater kit (LaMotte®). Catfish were given a two-week acclimation period before the trial began, with mortalities replaced with other fish. Following the adaptation period, fish were anesthetized with benzocaine (40 mg L^{-1}), weighed (g), and measured (fork length in cm), before being randomly assigned to one of six tanks (25 fish per tank) and divided into two groups, control and treated (fish exposed to formalin), with three replicates each. After stopping the water flow and decreasing the tank water volume (20 L) for 1 h, treated fish were given formalin (25 mg L^{-1}) baths twice a week; water flow was then restored, and tank water levels returned to normal. As a control, the fish were given a similar treatment but with water instead of formalin. During the treatments, the biofilters did not operate. To avoid regurgitation during anesthesia and obtain a more precise fish weight, fish were fed, with some changes, and according to Sánchez-Martínez et al. (2017), twice a day (09:00 and 15:00 h), six days a week, with a pelletized commercial 32% protein formula and fasted on the weighing day. Fish were sampled every two weeks for eight weeks following the initial measurement at the start of the trial (week 0). Fish were anesthetized, weighed (g), and individually measured (fork length, cm) on each sampling day. The amount of feed consumed per tank (g) was also recorded. The mean weight, condition index (K), specific growth rate (SGR), feed conversion ratio (FCR), and feed consumption (Rábago-Castro et al. 2006) were used to evaluate growth performance. After calculating growth performance indices for each tank, as well as the mean and standard deviations for the groups of tanks assigned to each treatment, statistical comparisons were made between the treated and control tanks every two weeks for the duration of the trial (0-8 weeks). The percentage of catfish mortality for each treatment was recorded at the end of the trial. A Student's *t*-test with a significance level $\alpha < 0.05$ was used to compare treated tanks to control tanks. Weekly mean weights of catfish were used for each group to fit growth curves using nonlinear least-squares, which were statistically compared using analysis of residual sum of squares (ARSS) (Haddon 2011).

RESULTS

During the experiment, water quality remained within optimal ranges for channel catfish. The mean (± standard deviation) temperature was 25.5 ± 0.17°C, which is within the favorable thermal window for growth and metabolic activity in this species. Dissolved oxygen averaged 6 ± 0.5 mg L^{-1}, consistently above the minimum acceptable threshold of 5 mg L^{-1}, ensuring adequate oxygen availability for respiration and minimizing physiological stress. The pH remained stable at 7.5 ± 0.6, which supports effective osmoregulation and minimizes the proportion of toxic un-ionized ammonia. Total ammonium (NH_3) concentrations averaged 0.2 ± 0.13 mg L^{-1}, indicating low levels of nitrogenous waste and efficient biofiltration, with calculated levels of un-ionized ammonia well below toxic thresholds.

There were no statistically significant differences ($P > 0.05$) between formalin-treated and control fish in K, FCR, or total feed consumption, indicating that formalin treatment did not adversely affect feed efficiency or general body condition. However, a significant difference ($P < 0.05$) in growth performance

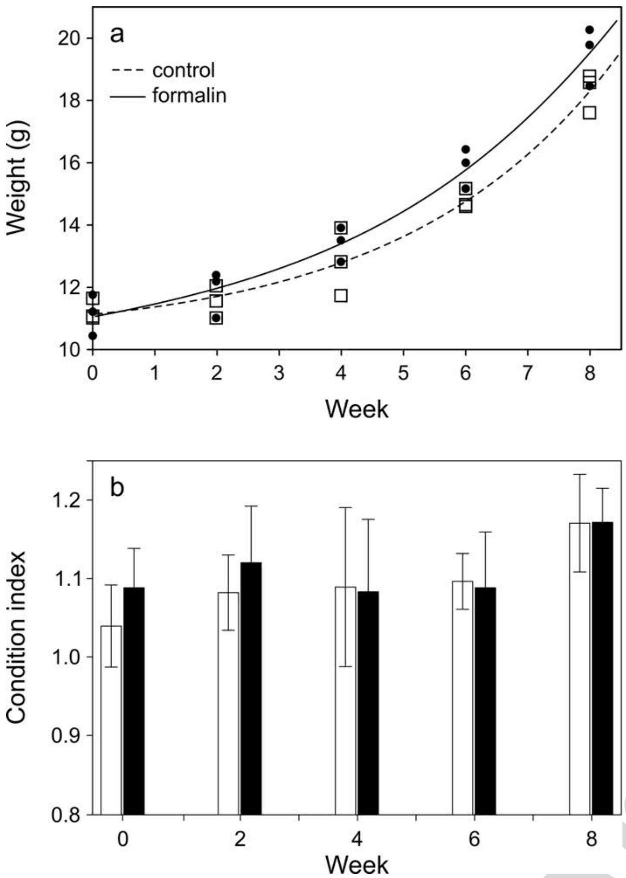

Figure 1. a) Growth in weight for channel catfish (*Ictalurus punctatus*). Control (blank squares) and formalin-treated (filled circles) groups during the trial. Fitted growth curves correspond to the equation $Y = a + b\,exp^{cX}$, which were significantly different (P < 0.05) from each other (F-statistics; Haddon 2011). b) The mean (± standard deviation) condition index (K) is also reported for both groups (control: blank columns, formalin: filled columns), which were not significantly different (P > 0.05) in any week.

was observed during the 0-8 week period. Formalin-treated fish exhibited a steeper growth trajectory, as shown in the growth curve (Fig. 1), and a significantly higher SGR compared to control fish (Table 1). No significant differences ($t_{(4)}$= 1.04, P = 0.36) in percentage of mortality were observed between the treated (24.9 ± 5.3) and untreated fish (21.2 ± 3.2). These findings suggest a potential growth-promoting effect of formalin under the tested conditions, possibly due to improved water hygiene or reduced microbial load. However, further investigation is needed to elucidate the underlying mechanisms.

Table 1. Mean (± standard deviation) specific growth rate (SGR; in % d^{-1}), feed consumption (in % of body weight), and feed conversion ratio (FCR) for channel catfish (*Ictalurus punctatus*) treated with formalin and untreated (control group) during the whole study period (0-8 weeks). The *P*-values for the comparison between groups are reported.

Parameter	Treatment		*P*
	Control	Formalin	
SGR (% d^{-1})	0.87 ± 0.06	1.00 ± 0.03	0.02
Feed consumption (% of body weight)	10.36 ± 0.71	10.29 ± 0.24	0.88
FCR	1.90 ± 0.18	1.65 ± 0.07	0.09

DISCUSSION

Formalin treatments against pathogens in cultured fish have been used to control and prevent diseases, highlighting the significance of understanding their potential effects on fish growth through bioassays. However, most of the studies done on the effects of formalin in fish do not focus on its effects on growth, but on other physiological variables (De Araújo et al. 2004, Leal et al. 2018, Hodkovicova et al. 2019, Tavares-Dias 2021), using doses and conditions that do not occur in commercial aquaculture.

Catfish exposed to prophylactic and intermittent use of formalin twice a week for eight weeks did not show significant differences in most of the growth indices examined, except for a significant increase in their SGR, which was reflected in a steeper growth curve observed in formalin-treated fish. The precise mechanism by which formalin significantly increased the SGR in fish in this study is unknown. However, one possible explanation is that because formalin reduces or eliminates external parasites and bacteria in fish gills, skin, and fins, avoiding subclinical infections, fish growth is promoted, as has been suggested in other animals such as pigs and poultry, where the use of antibiotics and chemotherapeutants in food is a common practice to promote growth (Jacela et al. 2009, Abed et al. 2019).

External formalin baths may alter gill mucous cells (Leal et al. 2018), leading to increased mucus production on the fish's skin. Consequently, pathogens entrapped in the mucus may be sloughed off (Dash et al. 2018). The use of formalin has been shown to increase the production and density of gill mucous cells in rainbow trout (*Oncorhynchus mykiss*; Buchmann et al. 2004). The effect of twice-weekly exposure to formalin (100 mg L^{-1}) on African catfish *Clarias* *gariepinus* fingerlings mean final weight, FCR and SGR were not significantly affected by formalin treatment after 10 weeks in a study by Jimoh et al. (2020); however, previously Jimmy et al. (2014) discovered that continuous doses of formalin reduce the growth of *C. gariepinus* fingerlings, which contradicts the findings of this study, in which the growth curve of threatened fish is significantly steeper than that of controls, and shows a trend where treated fish will grow more than controls.

Bodensteiner et al. (1993) discovered a significant improvement in K in channel catfish juveniles exposed to 25 ppm of formalin for consecutive days every week for 28 weeks, in contrast to our findings, which revealed no significant differences in K. Although formaldehyde is a highly toxic compound, its use (as formalin) is permitted in aquaculture (LaPatra & MacMillan 2008), and it quickly denatures in water; thus, its use may be similar to that of other chemical compounds, such as chloramine-T, which has been used as a prophylactic agent to prevent bacterial gill diseases in commercial salmonid hatcheries (Sanchez et al. 1996).

Exposure to formalin can disrupt osmoregulatory and respiratory functions in fish, potentially leading to mortality (Reardon & Harrel 1990). Nevertheless, in the current study, the formalin-treated fish did not exhibit a mortality rate significantly different from that of the untreated group. This outcome may be attributed to the relatively low formalin concentration employed in our study (25 ppm), which is considerably lower than the lethal concentrations documented for other freshwater fish species. For instance, the lethal concentration for *Cyprinus carpio* var. *koi* is 100 ppm (Tancredo et al. 2019), whereas for the African catfish *C. gariepinus*, it is approximately 90 ppm (Mbaru et al. 2011).

The effectiveness of formalin varies depending on the culture system, such as earthen ponds, raceways, and recirculation. Low-dose formalin does not appear to affect biofilter nitrification in recirculation systems, even when delivered continuously (Pedersen et al. 2010).

According to our findings, using formalin prophylactically at a low concentration of 25 ppm in intermittent baths improves the growth curve of channel catfish under experimental conditions. Future research with different marine or freshwater fish species and culture systems (such as intensive and recirculation ponds) would also be useful, as they affect chemical availability, degradation, and absorption.

CONCLUSIONS

During an eight-week trial, intermittent, prophylactic, low-dose (25 mg L^{-1}), 1-h, and biweekly formalin baths did not affect condition index, feed consumption, or feed conversion ratio in channel catfish (*I. punctatus*). However, formalin-treated fish had a significantly higher SGR and a steeper growth curve than control fish over the entire time interval (0-8 weeks). Under the conditions described in this study, formalin can be safely used prophylactically to prevent the onset of diseases in catfish, as it has no negative impact on growth and appears to improve it.

Credit the author's contribution

J.L. Rábago-Castro: conceptualization, methodology, review and editing; I.O. Montelongo-Alfaro: methodology, data collection; R. Pérez-Castañeda: methodology, validation, data curation and analysis, review, and editing; J.G. Sánchez-Martínez: funding acquisition, conceptualization, supervision, writing original draft, review and editing. All authors have read and accepted the published version of the manuscript.

Conflict of interest

The authors declare that there are no conflicts of interest.

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