



Short communication

Confirmation that pulse and continuous peracetic acid administration does not disrupt the acute stress response in rainbow trout



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ARTICLE INFO

Keywords:

Peracetic acid
Disinfection
Fish welfare
Stress

ABSTRACT

Peracetic acid (PAA) is considered an eco-friendly alternative to other antimicrobial agents of common use in aquaculture. The literature suggests that fish can habituate to PAA exposure based on a reduction of the fish corticosteroid response to PAA administration after repeated exposures. If that is true, PAA would also be a good option from the point of view of fish physiology. However, stronger evidence is needed to confirm that the use of PAA is welfare-friendly to fish. Besides habituation, other hypothetical factors such as desensitization, physiological exhaustion or PAA-mediated endocrine disruption could potentially explain the reduction in the corticosteroid response after repeated/prolonged PAA exposure. In this study, rainbow trout that had been exposed to PAA for several weeks were challenged with a secondary chasing stressor: fish were pursued with a dipnet for 1 min and their acute response was evaluated by measuring plasma cortisol, plasma glucose, plasma lactate and brain serotonergic activity. All fish were equally able to mount a normal physiological stress response to the secondary stressor independent of previous exposure to PAA. This suggests that the decrease in the cortisol response after repeated exposure to PAA, as seen in previous studies, is a true habituation to PAA administration, which supports the use of PAA as a welfare-friendly antimicrobial agent in aquaculture.

1. Introduction

Peracetic acid (PAA) has proven to be an efficient antimicrobial agent for aquaculture purposes (Meinelt et al., 2015; Liu et al., 2017a). In addition to its demonstrated effectiveness against fish pathogens (Farmer et al., 2013; Jussila et al., 2011; Smail et al., 2004; Lilley and Inglis, 1997), its degradation time and kinetics make it a good eco-friendly alternative to other disinfectants of common use in aquaculture such as formaldehyde, iodophors, phenolic compounds, chlorine or quaternary ammonium compounds (Danner and Merrill, 2005; Pedersen et al., 2009, 2013; Lahnsteiner and Kletzl, 2016; Liu et al., 2017a). Peracetic acid has been demonstrated to be acutely toxic to the following typical fish pathogens in vitro. Toxic concentrations of PAA were found to be $< 0.3 \text{ mg L}^{-1}$ against *Ichthyophthirius multifiliis* theronts, 0.8 mg L^{-1} against *I. multifiliis* tomites, 1 mg L^{-1} against *Flavobacterium columnare* and 4 mg L^{-1} against *Saprolegnia parasitica* (see Meinelt et al., 2007, 2009; Straus and Meinelt, 2009; Marchand et al., 2012). Recent research has shown that fish are able to tolerate PAA at low concentrations. The 24-h no observed effect concentration (NOEC)

for channel catfish (*Ictalurus punctatus*) yolk-sac fry was 2.2 mg L^{-1} PAA and 1.3 mg L^{-1} PAA for swim-up fry (Straus et al., 2012). The 24-h NOEC was $1.9\text{--}5.8 \text{ mg L}^{-1}$ PAA for a range of juvenile fish (Straus et al., in press). Several attempts of treating pathogens with PAA in the presence of fish were successful (Rintamaki-Kinnunen et al., 2005; Sudová et al., 2010; Jussila et al., 2011). To avoid the recurrence of pathogens, it is however necessary to use continuous or repeated exposures to PAA. In these cases, a welfare issue may emerge because the fish may suffer from chronic stress induced by the repeated exposures to PAA.

The stress response in common carp (*Cyprinus carpio*) and rainbow trout (*Oncorhynchus mykiss*) was recently examined when fish were exposed to repeated applications of PAA; this was done in an attempt to identify welfare-related issues regarding the use of PAA in aquaculture facilities (Liu et al., 2017a,b). In these studies, exposed fish exhibited an increase in the levels of plasma cortisol (the most common stress marker in fish) after the initial exposure to PAA, which indicated that the exposure was stressful. After repeated exposures to PAA, the cortisol response of the fish was lower, suggesting that the fish might have become habituated to the exposure, which would support the use of

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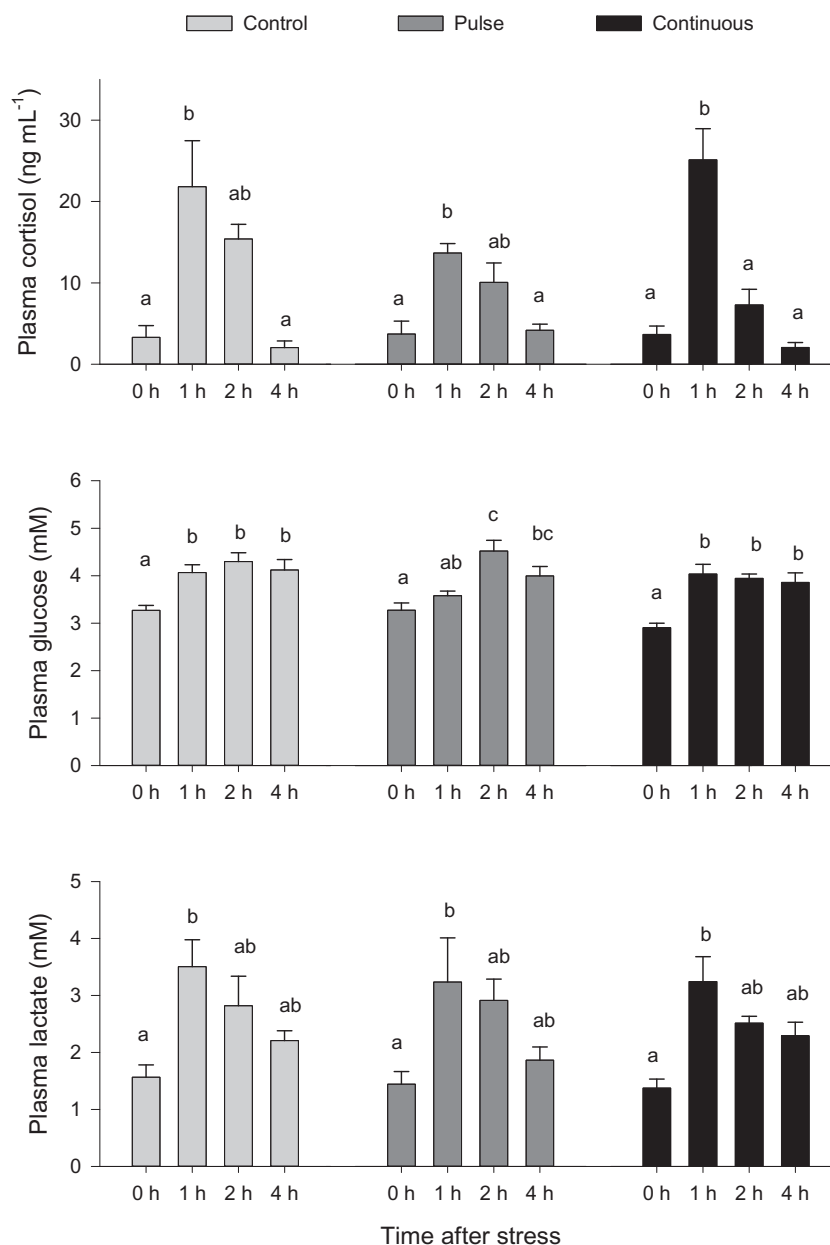


Fig. 1. Plasma levels of cortisol, glucose and lactate after 1 min of chasing stress in rainbow trout that were previously exposed to peracetic acid for 6 weeks. Columns represent the averaged values (and SEM) of $n = 6-8$ fish. Different letters represent statistically significant differences among time points for a given parameter and treatment group.

PAA as a welfare-friendly antimicrobial agent. However, lower levels of released cortisol are not necessarily the result of a process of habituation (Cyr and Romero, 2009). Alternative explanations to habituation for a cortisol response of smaller magnitude could be related to desensitization or exhaustion of the physiological stress response without habituation, or to potential PAA-induced alterations of the normal functioning of the hypothalamus-pituitary-interrenal (HPI) axis.

The present study determined whether these alternative explanations could be excluded, thus confirming that the fish are truly able to habituate to PAA exposure. According to our hypothesis, fish that are apparently habituated to PAA would be able to mount a normal physiological stress response (evaluated by measuring plasma cortisol, plasma glucose, plasma lactate and brain serotonergic activity) upon exposure to a different stressor.

2. Materials and methods

2.1. Fish, experimental design and sampling

Rainbow trout utilized during the study of Liu et al. (2017b) were used; their study evaluated how different types of PAA applications affected fish performance and system water quality in a flow-through aquaculture system. The applications were either repeated single dose (Pulse) or continuous via a peristaltic pump (Continuous). In brief, the following treatments were applied (in triplicate) to 180 L tanks containing 18 juvenile rainbow trout each: Control (no PAA exposure), Pulse (1 mg L^{-1} PAA, twice a week) and Continuous (0.2 mg L^{-1} PAA in the water inflow). This protocol was maintained for 6 weeks, after which the exposure experiment was completed. At that time, the average mass of the fish was 190.6 g ($\text{SD} = 29.9 \text{ g}$) with no differences among treatment groups (Liu et al., 2017b).

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