Aquacultural Engineering 63 (2014) 9–15

Contents lists available at ScienceDirect

Aquacultural Engineering

journal homepage: www.elsevier.com/locate/aqua-online

Evaluation of treatment methods using sodium percarbonate and formalin on Australian rainbow trout farms

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ARTICLE INFO

Article history:
Received 26 May 2014
Accepted 14 August 2014
Available online 16 September 2014

Keywords:
Sodium percarbonate
Formalin
Parasite management
Treatment distribution

ABSTRACT

In aquaculture, effective parasite management often relies on chemical therapy when prevention strategies are ineffective. Fish are often medicated using immersion (bath) treatments. The efficacy of bath treatment relies on maintaining at least the minimum effective dose for the necessary treatment duration. Dose is influenced by the product used, calculation of system volume, application method, flow, mixing, treatment degradation rate and environmental conditions. To maximize efficacy the relationships between system, delivery and effective dose need to be understood or controlled. We tested four application methods using sodium percarbonate (SPC) and two application methods using formalin (FOR, 37% formaldehyde [FA]) in four semi-closed flow-through systems on four Australian freshwater trout farms with different flow and water quality characteristics. Target dose was 64 mg/L SPC and 200 mg/L FOR. Hydrogen peroxide (HP) released from SPC was measured photometrically and FA levels were measured colorimetrically. Each application method achieved consistent doses across repeated applications but not all methods resulted in the dose reaching the target concentration in all parts of the system for the whole treatment duration. Eliminating the influence of system variables by creating static baths provides the most stable treatment environment. Where this is not possible, minimising system variables by modifying flow assists in retaining treatment in the system and improving accuracy of delivered doses. Treatment methods must be validated in a system prior to being routinely applied and mechanisms to optimise dose-duration identified and implemented.

1. Introduction

Rainbow trout, *Oncorhynchus mykiss* (Walbaum) are an ideal species for cultivation and are a key aquaculture species in Victoria, Australia (ABARES, 2012). One factor limiting productivity of the sector is ectoparasitic disease, primarily caused by *Ichthyophthirius multifiliis* (Fouquet). Outbreaks of *I. multifiliis* have a significant negative effect on the host (reviewed by Matthews, 2005), including mortality (Ewing and Kocan, 1992). Management of *I. multifiliis* largely centres on husbandry, including minimising stress, manipulating water velocity in raceways and, when these measures are inadequate, application of strategically timed chemotherapeutants.

Malachite green is an effective treatment for a wide range of ectoparasites but is a potential carcinogen and teratogen and is no longer permitted to be used in food fish aquaculture (Alderman, 1985; Wahli et al., 1993; Meinelt et al., 2009). Sodium percarbonate (SPC) and formalin (FOR) are viable alternatives to malachite green for treatment of ectoparasites (Heinecke and Buchmann, 2009). Sodium percarbonate is a granular solid that dissociates in water to release hydrogen peroxide (HP), a strong oxidising agent, which is active against ectoparasites over time (Noga, 2000; Heinecke and Buchmann, 2009). Sodium percarbonate can be used in Australia pursuant to a minor use permit (PER12944) issued by the Australian Pesticides and Veterinary Medicines Authority (APVMA) at doses of up to 100 mg/L, and is currently administered on Australian trout farms at a target dose of 64 mg/L (unpublished observations), which is effective against *I. multifiliis* theronts (Heinecke and Buchmann, 2009). FOR strongly reduces organic compounds (Masters, 2004), cross-links amino groups in proteins (Orlando et al., 1997) and is a general treatment for aquatic ectoparasites (Wise et al., 2004; Rowland et al., 2009). Formalin is typically administered at 200 mg/L for 1 h following Noga (2000) on Australian

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http://dx.doi.org/10.1016/j.aquaeng.2014.08.002
0144-8609/

Table 1
Description of the systems at Fish Farm A used in the validation of treatment methods using sodium percarbonate (SPC), measured as hydrogen peroxide (HP) and formalin (FOR) measured as formaldehyde (FA). N/A = not applicable, N/D = not determined.

Study system	SPC trials			FOR trials		
	Trial 1 Raceway 12	Trial 2 Raceway 6	Trial 3 Raceway 9	Trial 1 Raceway 12	Trial 2 Raceway 1	Trial 3 Raceway 2
Rearing volume (m^3)	55.8	33.6	34.3	55.8	34.3	57.4
Average depth (m)	1.2	0.72	0.73	1.2	0.73	1.25
In-flow water (L/s)	35	34	34	35	40	40
Fish density (kg/m^3)	32.2	N/D	N/D	32.2	26.8	14.4
Water temp. (°C)	6	5.2	5.4	5.5	9.7	9.8
COD (mg O_2/L)	<20	<20	<20	<20	<20	<20
Aeration	N/A	N/A	N/A	N/A	N/A	N/A
Treatment	SPC	SPC	SPC	FOR	FOR	FOR
Application method	Granular	Granular	Granular	Liquid	Liquid	Liquid
Initial dose	3.57 kg	2.15 kg	2.19 kg	8.04 L	6.8 L	11.48 L
Top ups	0.672 kg	0.65 kg	0.67 kg	2.01 L	2.4 L	2.4 L
Expected concentration of HP or FA (mg/L)	21	21	21	74	74	74

Table 2
Description of the flow-through systems at Fish Farm B used in the validation of liquid and granular application methods of sodium percarbonate (SPC), measured as hydrogen peroxide (HP). R = reduced flow, F = full flow, N/A = not applicable.

Study system	Liquid application trials			Granular application method		
	Trial 1	Trial 2	Trial 3	Trial 1	Trial 2	Trial 3
Type of system	Raceway 2	Raceway 4	Raceway 3	Raceway 4	Raceway 2	Raceway 1
Rearing volume (m^3)	23.2	17.4	21.6	17.4	23.2	18.4
Average depth (m)	0.5	0.5	0.5	0.5	0.5	0.5
In-flow water (L/s)	R 6.4, F 17.5	R 9.8, F 11.8	R 10.3, F 12.7	9.8	10.4	10.3
Fish density (kg/m^3)	29	10.6	13.9	24	16.1	20.2
Water temp. (°C)	6.4	11.2	11.3	6.8	11.4	11.5
COD level (mg O_2/L)	29	<20	<20	<20	<20	<20
Aeration	N/A	N/A	N/A	N/A	N/A	N/A
Treatment	SPC	SPC	SPC	SPC	SPC	SPC
Initial dose	1.152	1.152	1.152	1.057	1.482	1.179
Top ups (kg)	0.64	0.64	0.64	0.903	0.148	0.117
Expected concentration of HP (mg/L)	21	21	21	21	21	21

trout farms, which is effective against *I. multifiliis* (see Wahli et al., 1993; Lahnsteiner and Weismann, 2007; Heinecke and Buchmann, 2009).

In Australia, rainbow trout are typically cultured in flow-through concrete raceways or earthen ponds, where FOR and SPC have been reported to be ineffective against *I. multifiliis* (pers. comm. E. Meggit, Victorian Trout Grower's Association). The major influences on the efficacy of bath treatments are the distribution of the product in the system and achieving and maintaining the minimum effective concentration for the treatment period (Rach et al., 1997). Under-dosing and uneven distribution of the product in the system is therefore the most likely cause of low efficacy. The distribution of the dose in aquaculture systems is affected by application method, flow, mixing, degradation rate, environmental and water conditions (Rach and Ramsay, 2000). Understanding how these conditions influence the treatment is critical for achieving the target dose; if one of these variables is altered it may lead to the dose being too low or the exposure too short for appropriate efficacy, or too high or prolonged, with potentially negative effects on the exposed fish.

We designed this study to evaluate FOR and SPC application methods used on four commercial trout farms to determine if the minimum effective dose and desired duration were being achieved throughout each system.

2. Materials and methods

2.1. Field trials

Treatment application methods were assessed in four semi-closed flow-through systems on four fish farms with different flow and water quality characteristics: Fish Farm A, a concrete, low volume, high turnover flow-though system; Fish Farm B, a concrete, low volume, low turnover flow-through system; Fish Farm C, a concrete, high volume, low turnover, flow-through system; and Fish Farm D, a low turnover, high volume, flow-through earthen raceway. Flow-though systems on each farm had variable characteristics and were stocked with 1-year old juvenile rainbow trout at different densities, outlined in Tables 1–4. In all experiments, treatments were applied using the standard method used on that farm based on the volume of the system and the residual active compound was measured (SPC was measured as HP; FOR was measured

Table 3
Description of the flow-though systems at Fish Farm C used in the validation of application methods of formalin (FOR) measured as formaldehyde (FA).

Study system	Fish Farm C		
	Trial 1	Trial 2	Trial 3
Type of system	Pond 2	Pond 5	Pond 3
Rearing volume (m^3)	191	172	152
Average depth (m)	1.3	1.2	1.05
In-flow water (L/s)	7.82	7.82	10.08
Fish density (kg/m^3)	35.6	33.5	30
Water temp. (°C)	6.0	6.1	10.4
COD level (mg O_2/L)	<20	<20	<20
Aeration	Paddle wheel	Paddle wheel	Paddle wheel
Treatment	FOR	FOR	FOR
Application method	Liquid	Liquid	Liquid
Initial dose	20 L	20 L	20 L
Top ups (kg)	–	–	–
Expected concentration of FA (mg/L)	74	74	74