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Safety and efficacy of the Environmental Products Group Masterflow Aquarium Management System with Aegis Microbe Shield™

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Abstract

This study investigated the safety and efficacy of the EPG Masterflow Aquarium Management System with Aegis Microbe Shield™ (EPG-MAMS). Four different species of fish were used in the study. Ten fish of each species were placed in 75 l aquariums containing the EPG filter media, a commercially available filter media (Whisper®) and an aquarium with no filter material. At the end of the 45 day trial three fish from each tank were sacrificed and preserved in formalin for histopathology. Water quality parameters were routinely monitored. The EPG filter media was compared with the Whisper® filter media for efficacy against *Aeromonas salmonicida* using a shaker flask microbiological assay. The EPG filter proved to be clinically and histopathologically safe and reduced to some degree the number of *A. salmonicida* suspended in water in an in vitro study. © 1999 Elsevier Science B.V. All rights reserved.

Keywords: Environmental products group; Masterflow aquarium management system; Aegis microbe shield

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1. Introduction

Recent statistics compiled by the Pet Industry Joint Advisory Council (PIJAC, 1997) indicate that 10% of US households (9.4 million) maintain freshwater aquaria with an additional 0.6 million households containing marine aquaria. The PIJAC stated that ‘aquarium treatments, remedies, medications and water conditioners are sold by approximately 11000–14000 pet stores and aquarium stores; 4000–5000 discount stores; 75000 grocery stores, food warehouses, variety stores, drug stores, garden centers, feed stores and other retail outlets.

Water borne viral, bacterial, fungal, protozoal, helminth and crustacean pathogens plague the ornamental fish industry. The two most effective and commonly employed methods for disinfecting recirculating aquatic systems designed for fin fish and invertebrates are ozonation and ultraviolet (UV) light sterilization (Colberg and Lingg, 1978; Rosenthal, 1981; Paller and Lewis, 1988). Efficacy of these systems is complex and depends upon a number of variables including the particular mechanical system employed, flow rate, contact time, targeted pathogens and species of fish in the system. Additionally, ozonation presents certain risks to the health of the fish and humans in contact with the system (Wedemeyer et al., 1979; Rosenthal, 1981). Ozonation and UV sterilization are also relatively expensive systems to install and operate and as such, are not practical for many home aquariums.

A filter with bacteriocidal activity that is also safe for pet fish is highly desirable and could possibly reduce the practice of indiscriminate ‘shotgun’ antimicrobial usage. Such a filter, if safe for target and non-target species, would also have potential value to recirculating food fish and invertebrate aquaculture operations.

This study was undertaken to determine whether or not the EPG-MAMS is safe for four species of representative freshwater ornamental fish species during a 45 day trial and to determine whether the EPG-MAMS reduces the water content of an important bacterial pathogen of freshwater fish.

The EPG-MAMS employs a high density synthetic blend mesh which has been impregnated with the Aegis Microbe Shield™, a patented and EPA registered (No. 34292-1) compound which possesses a unique surface-active chemistry that purportedly destroys airborne bacteria, fungi and algae.

2. Materials and methods

Four different species of fish were used for this study: lemon tetra (*Hemigrammus boesmani*), the venustus African cichlid (*Haplochromis venustus*), the common black mollie (*Poecilia sphenops*) and the medaka (*Oryzias latipes*). All fish were captive reared and were acclimated and quarantined for 30 days before being placed in the experimental and control aquaria. On day 1 of the 45 day safety trial 3 specimens from each species were humanely euthanized with an

overdose of tricaine methanesulfonate (300 mg/l for 10 min) and fixed in 10% neutral buffered formalin for histopathological examination. These fish served as histopathological controls.

Aquarium systems consisted of a 75 l tank, 75 W submersible heater, Whisper II® power filter and a floating thermometer. A total of 12 aquaria were included in the safety trial. Ten specimens of each of the 4 species were placed in aquaria containing either the EPG-MAMS filter cartridge, Whisper® cartridge or no cartridge at all (these aquaria served as the true controls). Aquaria were monitored daily for mortality and water temperature and pertinent water quality parameters (temperature, pH, ammonia, nitrite and nitrate) were monitored three times weekly. Fish were fed once every 24 h a consistent volume of commercial tropical fish food. At the end of the 45 day trial three fish from each aquarium were humanely euthanized with an overdose of tricaine methanesulfonate (300 mg/l for 10 min) and fixed in 10% neutral buffered formalin for histopathological examination.

Aeromonas salmonicida is an important pathogen of fishes and is typically treated with a variety of antimicrobial compounds (Barnes et al., 1990; Inglis and Richards, 1991; Martinsen et al., 1992; Stoffregen et al., 1993). The in-vitro 'shaker flask' method was used to test the in vitro antimicrobial activity of the EPGMAMS using the modifications needed to accommodate *A. salmonicida* (Dow Corning Report, 1979).

3. Results

Mortality levels were low in all 12 aquariums and there was no statistical difference between experimental and control filtration systems (Fig. 1). The EPG filter produced no abnormal results and, in some aquaria, appeared more efficient at reducing ammonia and nitrite levels.

Histopathological examination of 48 fishes (12 pre experimental controls and 36 post experimental specimens) showed no microscopic changes in the following tissues: gill, skin, gastrointestinal tract, heart, kidney, liver, pancreas, spleen, striated muscle. There was no evidence of any toxic changes or toxicity induced necrosis in any organs. Several of the control (pre trial) and experimental African cichlids had large granulomas of the intestinal wall.

In two separate in vitro trials the EPG filter out performed the Whisper® filter in removing bacteria from a 100 ml water sample. In the first trial (Table 1), the mean number of bacteria (three replicates) decreased from 3.02 to 2.07 bacteria/ml (paired *t*-test value of 0.05) after 1 h of incubation in the shaker flask with the EPG sample (s) while the Whisper filter sample (c) showed an increase from 2.31 to 3.02 bacteria/ml (*t*-test value = 0.14). In the second trial (Table 1), the EPG-MAMS filter outperformed the Whisper® filter, however, both samples showed gains in bacteria/ml after the 1 h incubation period. The mean values for the EPG-MAMS sample rose from 3.05 to 3.51 (*t*-test value = 0.18) while the Whisper® values increased from 2.57 to 3.70 (*t*-test value = 0.01).

4. Discussion

The results of this study show that the EPG-MAMS is safe for a variety of tropical fish species and does not produce clinical disease or microscopic lesions. The EPG filter did not have any adverse affects on pH or other water quality parameters. Since ammonia and nitrite levels were equivalent to or below levels

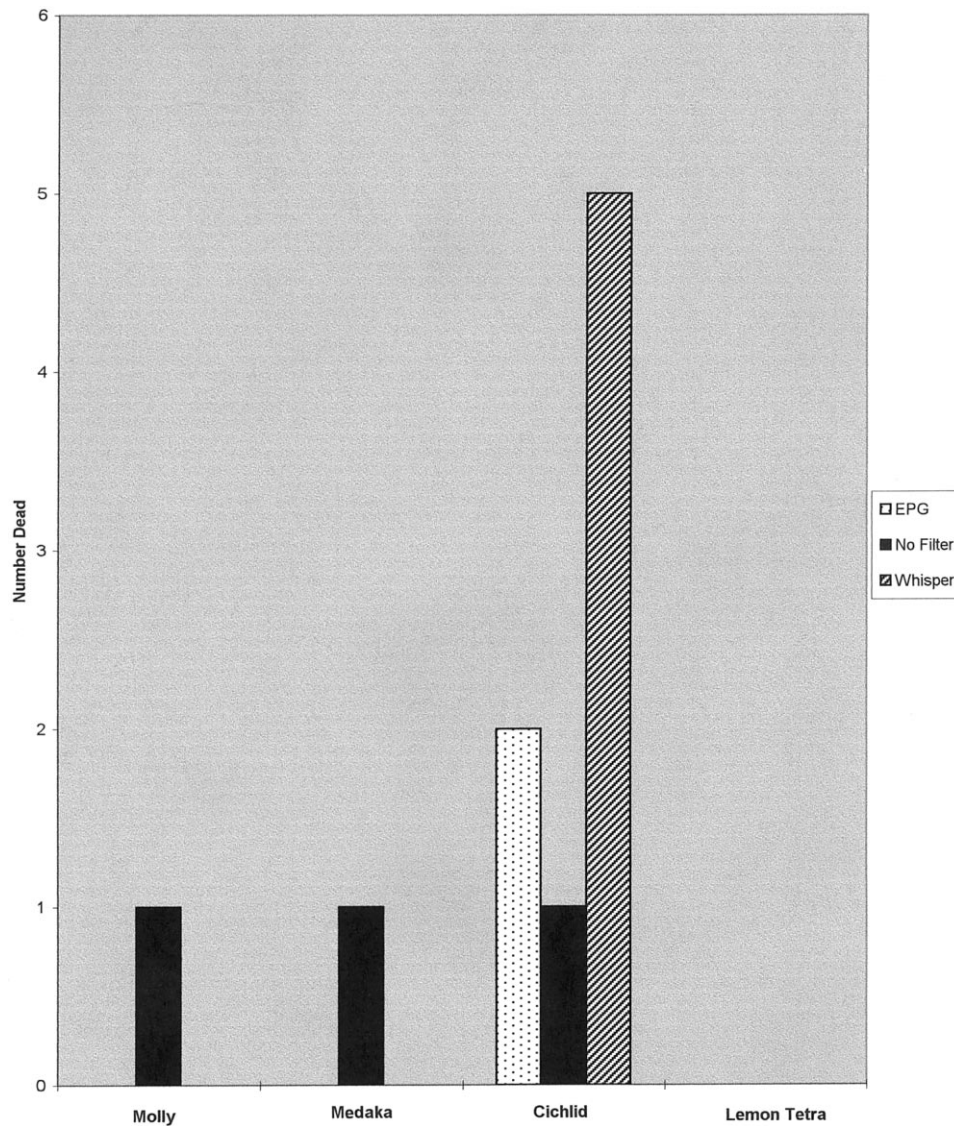


Fig. 1. Mortality for the four species of fish tested. There was no mortality among the lemon tetras.

Table 1
Mean bacterial counts (number/ml) at time zero and 1 h following flask trial (standard deviations follow in [])

Trial no.	Time (h)	Control	EPG
1	0	2.31 [0.90]	3.02 [0.025]
	1	3.02 [0.41]	2.07 [0.76]
2	0	2.57 [1.32]	3.05 [0.28]
	1	3.70 [1.52]	3.51 [0.30]

Control values are for the Whisper[®] filter pad.

recorded for the control aquaria, it appears that the EPG-MAMS material did not have a deleterious effect on nitrifying bacteria.

The in vitro antimicrobial results show that the EPG filter does limit replication of *A. salmonicida* in culture media and in one trial reduced bacterial numbers after a 1 h exposure. The experimental results were statistically significant using a paired *t*-test in the first trial, while the control results were not statistically significant in the same trial. In the second trial, the control results were statistically significant, while the experimental results were not. The Whisper filter material (which does not claim to have antimicrobial activity) used as a control did not limit bacterial replication and numbers of bacteria increased in all trials.

The EPG-MAMS reduced to some degree the number of *A. salmonicida* suspended in water, but further research is needed to evaluate its long-term efficacy.

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