Florfenicol Medicated Feed Clinical Field Trials - INAD 10-697

Year 2005 Annual Summary Report on the Use of Florfenicol Medicated Feed in Field Efficacy Trials

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Summary

Florfenicol-medicated feed (FMF) has been used effectively in the U.S. under compassionate INAD Exemption #10-697 to control mortality in a variety of fish caused by common fish bacterial pathogens. In calendar year 2005 the efficacy of FMF was evaluated in 74 disease trials involving approximately 10.9 million fish to control mortality in a variety of test fish caused by a variety of infectious fish pathogens. Trials were conducted at a total of 13 fish culture facilities, including three U.S. Fish and Wildlife Service National Fish Hatcheries (NFH), and two state, four private, and four tribal fish hatcheries. Florfenicol medicated feed was administered at the standard treatment dose of 10 mg/Kg fish/d for 10 d in 73 studies. In the remaining trial, the Investigator deviated from the protocol and administered feed at dose of 15 mg/kg for 10 d. Overall results indicated that treatment appeared effective in approximately 82% of the trials, and was characterized as inconclusive in the remaining trials.

Introduction

Bacterial diseases are a major problem in aquaculture and account for significant losses of fish (Clarke and Scott 1989; Frerichs and Roberts 1989; Bjorndal 1990). Although the importance of environmental conditions (McCarthy and Roberts 1980; Haastein 1988; Munro and Roberts 1989) and the value of effective vaccines, where available (Ellis 1989), are acknowledged, antimicrobial therapy presently has an important role to play in aquaculture (Klontz 1987; Alderman 1988). Florfenicol is a potent, broad-spectrum, antimicrobial agent with bacteriostatic properties (Horsberg et al. 1996). It is a fluorinated analogue of thiamphenicol and is also similar in structure to chloramphenicol, both of which have been used as broad-spectrum, veterinary antibiotics (Nagata and Oka 1996).

Florfenicol has great potential for treatment of infectious diseases, and because of its high potency and safety to humans, it could become an important drug in veterinary medicine, especially with respect to animals used by humans for food (Powers et al. 1990). Additionally, because florfenicol is not currently used in human medicine, it has become a strong candidate for use in aquaculture, and there is considerable interest to obtain U.S. Food and Drug Administration (FDA) approval for its use in fish culture.

The proposed treatment strategy (i.e., dosage and duration) for the use of FMF in fish is designed to meet the needs of individual fish species, individual fish lots, and a

variety of environmental conditions. In all cases, treatment goals are to (1) minimize the negative effects of disease on fish health, quality, and survival, and (2) help meet fishery management objectives. Because many factors can affect the success or failure of florfenicol-medicated feed therapy, supplemental efficacy data from compassionate Investigational New Animal Drug (INAD) use, as well as efficacy data from controlled, replicated studies that are scientifically valid and statistically defensible (i.e., pivotal), are needed to gain approval of FMF use in aquaculture.

Purpose of Report

The purpose of this report is to summarize the results of supplemental FMF field efficacy studies conducted in calender year (CY) 2005. Furthermore, it is expected that these data will be used to enhance the FMF database for the purpose of developing an appropriate label claim for the use of this new animal drug.

Facilities, Materials, and Treatment Procedures

1. Participating Facilities

A total of 74 effectiveness trials were conducted at 13 fish culture facilities, including three U. S. Fish and Wildlife Service (Service) National Fish Hatcheries (NFH), and two state, four private, and four tribal fish hatcheries. Trials were conducted to control mortality in a variety of fish caused by a variety of fish

pathogens. Water temperature during treatment trials ranged from 41.6 - 81.0 °F, with a mean treatment temperature of 71.5 °F.

2. FMF used in trials

The Aquaflor™ used in CY 2005 trials contained 500 g of florfenicol per kg of premix. Florfenicol is a pure compound with no inactive ingredients. All florfenicol used was supplied as Aquaflor™ by Schering-Plough Animal Health, 1095 Morris Avenue, Union, NJ. Florfenicol medicated feed was prepared by either top-coating florfenicol onto commercial fish feed at the Bozeman Fish Technology Center using a Standard Operating Procedure developed by the Services Aquatic Animal Drug Approval Partnership Program, prepared at the testing site by the Investigator, Monitor, or their designee, or prepared by commercial fish feed manufactures.

3. Drug dosages and duration

As described in the Study Protocol for INAD #10-697, Investigators were allowed to use FMF at 10 mg of active drug/kg of fish/d for 10 d. However, in one trial one group of test fish were fed FMF medicated feed at 15 mg/Kg fish/d for 10 d as part of a side-by-side study to compare results from a group of test fish treated with 10 mg/Kg fish/d for 10 d (please see Study Number 10-697-05-15 for more details). Due to fish size (1.85 g), the inherent withdrawal period for the tested fish will be approximately 300 d after treatment (i.e., fish will not be

available for human consumption until they grown to a sufficient size, which was projected at 300 d).

Fish Species and Fish Diseases Involved in CY 2005 Trials

1. Species of fish treated

Six fish species, including four species of salmonids and two non-salmonid fish species, listed below were treated with FMF during CY 2005. Treated fish ranged in size from 1.2 - 11.8 in.:

Salmonids:

coho salmon *Oncorhychus kisutch*

cutthroat trout *O. clarki*

rainbow trout *O. mykiss*

steelhead trout *O. mykiss*

Non-salmonids:

hybrid striped bass *Morone americana* x *M. saxatilis*

largemouth bass *Micropterus salmoides*

2. Diseases treated

Test fish diagnosed with one of the following diseases were treated during the reporting period: (1) Aeromonas liquefaciens, (2) Aeromonus/pseudomonas, (3)

bacterial coldwater disease, (4) columnaris, (5) furunculosis, (6) general systemic bacterial infection, (7) motile Aeromonad, or (8) streptococcal septicemia.

Data Collected

1. Pathologist's reports

Pathologists reports were submitted with 16 of the 74 trials conducted during CY 2005. Fish health pathology reports included: 1) a description of how the identity of disease agent(s) was verified; 2) disease identification records that confirm the presence of the disease agent; and 3) the name and title of the individual performing the diagnosis. Additionally, pathology reports often provide documentation that there were no secondary infections or infestations caused by unrelated disease agents in the population of test fish. Pathology reports provide critical information if such submissions are to be used in support of an initial approval, or to expand/extend an existing approved label.

2. Mortality data

As stated in the Study Protocol, mortality data were to be collected 10 days prior to treatment, during the treatment period, and for at least 21 days post-treatment. Investigators were strongly encouraged to collect mortality data on a daily basis. However, daily collection of pre-treatment mortality data was not always possible due to fish being moved (i.e., split into additional rearing units, or combined with fish from another rearing unit) from rearing unit to rearing unit.

Discussion of Study Results:

1. General observations on the efficacy of FMF for the control of bacterial diseases in salmonid and non-salmonid fish (Note: Table 1 provides a summary of all efficacious trials; Table 2 provides a summary of all inconclusive trials; Table 3 provides general CY 2005 summary data; and Table 4 provides a brief description of all trials conducted during CY 2005 under INAD #10-697.)

A. Efficacy at 10 mg/Kg fish/d for 10 days under INAD #10-697

Coho salmon, cutthroat, rainbow, and steelhead trout, hybrid striped bass, and largemouth bass were treated with 10 mg florfenicol/Kg fish/d for 10 days in 73 trials to control mortality caused by one of the previously described fish diseases (Tables 1 - 2). FMF treatments appeared effective in 60 (82%) of the 73 trials; whereas treatments in the remaining 13 (18%) trials were characterized as inconclusive.

B. Efficacy at 15 mg/Kg fish/d for 10 days under INAD #10-697

Steelhead trout were treated with 15 mg florfenicol/Kg fish/d for 10 days in one trial to control mortality caused by bacterial coldwater disease (Table 1). FMF treatment in this study appeared effective.

2. Observed Toxicity

No toxicity or adverse effects relating to FMF treatment were reported.

Summary of Study Results

Florfenicol medicated feed was administered to test fish in 74 separate trials at a dosage of either 10 or 15 mg/Kg fish/d for 10 d. Six different fish species were treated with FMF, and trials involved approximately 10.9 million fish. Treated fish ranged in size from 1.2 - 11.8 in. Water temperature during treatment ranged from 41.6 - 81.0 °F, with a mean treatment temperature of 71.5 °F. Overall results showed that in approximately 82% of the trials, FMF treatments appeared effective, and in 18% of the trials, treatment results were characterized as inconclusive. Data from these studies will be considered as ancillary data because of a general lack of quality control criteria essential for pivotal or supportive studies, such as use of untreated control fish, dose verification, replication, and randomization. None-the-less, the ancillary data documented in this report should provide useful corroborative data to support a new label claim for FMF. It is anticipated that additional ancillary efficacy data will continue to be collected under INAD #10-697. In future trials conducted under INAD #10-697, efforts will be directed towards the generation of higher quality data.

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Table 1. Summary of CY 2005 Florfenicol Medicated Feed Efficacy Results - Effective Trials

Hatchery	Number of Trials	Fish Species	Fish Size (inches)	Number of Fish	Disease	Number of Treatment Days	Dose (mg/kg)	Temp. (°F)
Lower Elwha	2	COS	1.3 - 1.9	1,018,877	CWD	10	10	49.0
Makah NFH	3	COS	3.5 - 4.0	629,618	Furunculosis	10	10	58.3 -63.1
Nez Perce Tribal Hatchery	1	COS	6.00	289,535	CWD	10	10	41.6
Prosser Hatchery	1	COS	4.40	115,000	Furunculosis	10	10	43.0
Whitman Lake Hatchery	1	COS	3.20	242,506	A. liquefaciens	10	10	53.4
Glenwood Sport SFH	1	CUT	1.15	137,227	CWD	10	10	58.0
St. Croix Waters Fishery	1	LMB	8.00	8,300	A./pseudomonas	10	10	72.7
Benner Spring SFH	2	RBT	3.0 - 4.0	222,000	CWD	10	10	50.0
Glenwood Sport SFH	1	RBT	3.59	192,893	CWD	10	10	58.0
Harrietta Hills Trout Farm	1	RBT	5.00	59,836	Motile Aeromonad	10	10	57.5
Tellico Enterprises	2	RBT	2.0 - 3.6	3,360,000	CWD	10	10	42.9 - 57.0
Dworshak NFH	2	STT	4.02	531,573	CWD	10	10	45.4 - 45.9
Makah NFH	1	STT	1.90	138,013	CWD	10	10	55.4
Tionesta FCS	1	STT	3.00	703,000	CWD	10	10	67.3
Kent SeaTech Corp.	3	SXW	1.20	137,446	Columnaris	10	10	79.0
	37	SXW	1.2 - 11.8	1,272,164	Streptococcus	10	10	79.0 - 81.0
Makah NFH	1	STT	2.30	62,081	CWD	10	15	55.4

Table 2. Summary of CY 2005 Florfenicol Medicated Feed Efficacy Results - Inconclusive Trials

Hatchery	Number of Trials	Fish Species	Fish Size (inches)	Number of Fish	Disease	Number of Treatment Days	Dose (mg/kg)	Temp. (°F)
Lower Elwha Hatchery	1	COS	2.50	917,551	CWD	10	10	49.0
Whitman Lake Hatchery	1	COS	3.20	238,662	A. liquefaciens	10	10	53.4
Harrietta Hills Trout Farm	1	RBT	2.50	187,096	General Systemic Bacterial Infection	10	10	48.0
	1	RBT	5.00	154,311	Motile Aeromonad	10	10	48.0
Kent SeaTech Corp.	9	SXW	1.2 - 11.8	234,219	Streptococcus	10	10	79.0 - 81.0

Table 3. Summary Data Regarding CY 2005 Florfenicol Medicated Feed Efficacy Trials

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Total Fish Treated:		**10,851,908**
Number of fish treated in effective trials		9,120,069
Number of fish treated in inconclusive trials		1,731,839
Total number of trials:		**74**
Number of effective trials:		61
Number of inconclusive trials:		13
Treatment Regimes Used:		
10mg/Kg fish/day for 10 days		73 trials
15mg/Kg fish/day for 10 days		1 trial
Treatment Water Temperature (°F):		
Temperature Range	41.6 - 81.0	
Mean Temperature	71.5	
Size of Treated Fish (in.):		
Size Range	1.15 - 11.8	

Species Treated:

coho salmon *Oncorhychus kisutch*
cutthroat trout *O. clarki*
rainbow trout *O. mykiss*
steelhead trout *O. mykiss*
largemouth bass *Micropterus salmoides*
hybrid striped bass *Morone americana* x *M. saxatilis*