Florfenicol Medicated Feed Clinical Field Trials - INAD 10-697

Year 2002 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 (CY) 2002 the efficacy of FMF was evaluated in 22 disease trials involving 898,325 fish to control mortality in a variety of test fish caused by bacterial coldwater disease, columnaris, furunculosis, pseudomonas aeromonas, or streptococcal septicemia. Trials were conducted at three U.S. Fish and Wildlife Service National Fish Hatcheries (NFH), four state, one private , and one tribal fish hatchery. All FMF was administered at the standard treatment regimen of 10mg/Kg fish/d for 9 - 11 d. Overall results of trials conducted in CY 2002 indicated that approximately 64% of the trials appeared efficacious, 13% appeared ineffective, and 23% were characterized as inconclusive.

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 primary purpose of this report is to summarize the results of CY 2002 supplemental FMF field efficacy studies. However, it is also 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 3 National fish hatcheries, 4 state, 1 private, and 1 tribal fish hatchery (n = 9 fish hatcheries) used FMF to control mortality in a variety of freshwater fish caused by bacterial coldwater disease, columnaris, furunculosis, Pseudomonas

aeromonas, or streptococcal septicemia. Water temperature during treatment trials ranged from 46.6 - 86.0 °F, with a mean treatment temperature of 70.4 °F.

2. FMF used in trials

The Aquaflor used in CY 2002 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 Aqauflor 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 accepted Standard Operating Procedures, 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 three trials fish were fed FMF medicated feed at 10 mg/Kg fish/d for either 9 or 11d.

Fish Species and Fish Diseases Involved in year 2002 Trials

1. Species of fish treated

The three salmonid species and four non-salmonid fish species listed below were treated with FMF during CY 2002. Treated fish ranged in size from 1.0 - 9.3 in. :

Salmonids:

coho salmon *Oncorhynchus kisutch*

cutthroat trout *O. clarki*

steelhead trout *O. mykiss*

Non-salmonids:

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

muskellunge *Esox masquinongy*

tiger muskellunge *E. lucius x E. masquinongy*

yellow perch *Perca flavescens*

2. Diseases treated

Test fish diagnosed with one of the following diseases were treated during the reporting period: (1) bacterial coldwater disease, (2) columnaris, (3) furunculosis, (4) pseudomonas aeromonas, or (5) streptococcal septicemia.

Data Collected

1. Pathologist's reports

Pathologists reports were submitted with 14 of the 22 trials conducted during CY 2002 (7 of the 14 trials in which pathology reports were submitted were pivotal studies and comprehensive pathology reports were included with the pivotal data submissions). 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 was 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 non-efficacious trials; Table 3 provides a summary of all inconclusive trials; Table 4 provides general CY 2002 summary data; and Table 5 provides a summary of all trials conducted during CY 2002 under INAD #10-697.)

A. Efficacy at 10 mg/Kg fish/d for 9 - 11 days under INAD #10-697

FMF was used at 10 mg/Kg fish/d for 9 - 11 days in 15 trials in which the test fish were hybrid striped bass, muskellunge, steelhead trout, tiger musky, and yellow perch diagnosed with either bacterial coldwater disease, columnaris, pseudomonas aeromonas, or streptococcal septicemia (Tables 1 - 3). FMF treatment appeared efficacious in 8 trials, while 2 trials were inefficacious, and 5 trials were characterized as inconclusive.

B. Efficacy at 10mg/Kg fish/day for 10 days under Protocol FLOR-01-EFF

FMF was used at 10 mg/Kg fish/d for 10 days in seven trials in which the test fish were coho salmon, cutthroat trout, steelhead trout, and hybrid striped bass diagnosed with either furunculosis, CWD, columnaris, or streptococcal septicemia (Tables 1 - 2). FMF treatment was effective in 6 trials, and ineffective in 1 trial (please refer to pivotal submission numbers FLOR-01-

EFF-05, FLOR-01-EFF-06, FLOR-01-EFF-08, FLOR-01-EFF-09, FLOR-01-EFF-12, FLOR-01-EFF-13, and FLOR-01-EFF-13B for more details on these studies).

2. Observed Toxicity

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

Summary of Study Results

Florfenicol medicated feed was used at 10 mg/Kg fish/d. Treatment duration ranged from 9 - 11 days. Seven different fish species were treated with FMF, and trials involved 898,325 fish. Treated fish ranged in size from 1.0 - 9.3 in. Water temperature during treatment ranged from 46.6 - 86.0 °F, with a mean treatment temperature of 70.4 °F. Overall results showed that approximately 64% of the trials appeared efficacious, 13% appeared ineffective, and 23% were characterized as inconclusive. Trials conducted under the research protocol FLOR-01-EFF included use of control fish, detailed pathologist's reports documenting the disease during the trials, and will likely be accepted by CVM as pivotal or supportive. Data from the other studies can only 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, 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.

References

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Table 1. Summary of CY 2002 Florfenicol Medicated Feed Efficacy Results - Efficacious Trials

Hatchery	Number of Trials	Fish Species	Fish Size (inches)	Number of Fish	Disease	Number of Treatment Days	Dose (mg/kg)	Temp. (°F)
Bellingham SFH	1	COS	3.50	24,822	Columnaris	10	10	69.4
Makah NFH	1	COS	3.50	1,200	Furunculosis	10	10	63.1
Washoe Park Trout SFH	1	CUT	1.00	4,127	CWD	10	10	48.7
Kent SeaTech Corp.	6	SXW	4.3 - 7.4	150,180	Steptococcus	10	10	82.4 - 86.0
Makah NFH	2	STT	2.3 - 2.4	159,320	CWD	10 - 11	10	57.0 - 57.5
St.Croix Waters Fishery	3	YEP	3.4 - 5.7	173,573	Columnaris & Pseudomonas Aeromonas	10 - 11	10	71.0

Table 2. Summary of CY 2002 Florfenicol Medicated Feed Efficacy Results - In-Efficacious Trials

Hatchery	Number of Trials	Fish Species	Fish Size (inches)	Number of Fish	Disease	Number of Treatment Days	Dose (mg/kg)	Temp. (°F)
Bozeman FTC	1	CUT	2.20	900	CWD	10	10	55.8
Benner Springs Fish Research	1	MUH	5.00	4,000	Columnaris	10	10	66.2
St.Croix Waters Fishery	1	YEP	8.40	8,170	Columnaris & Pseudomonas Aeromonas	10	10	71.0

Table 3. Summary of CY 2002 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)
Spirit Lake SFH	1	MUE	1.93	3,000	Columnaris	10	10	69.8
Kent SeaTech Corp	1	SXW	4.25	125,851	Streptococcus	10	10	86.0
Dworshak NFH	1	STT	4.30	50,000	CWD	9	10	46.6
Makah NFH	1	STT	2.20	65,000	CWD	10	10	62.0
St.Croix Waters Fishery	1	YEP	9.30	128,182	Columnaris & Pseudomonas Aeromonas	10	10	71.0

Table 4. Summary Data Regarding CY 2002 Florfenicol Medicated Feed Efficacy Trials

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Total Fish Treated:	**898,325**
Number of fish treated in efficacious trials	513.222
Number of fish treated in inefficacious trials	13,070
Number of fish treated in inconclusive trials	372,033
Total number of trials:	**22**
Number of efficacious trials:	14
Number of inefficacious trials:	3
Number of inconclusive trials:	5

Pivotal Studies:

Study Number: 10-697-02-4; 10-697-02-5; 10-697-02-7; 10-697-02-8; 10-697-02-10; 10-697-02-11; 10-697-02-12

Treatment Regimes Used:

10mg/Kg fish/day for 9 - 11 days	22 trials

Treatment Water Temperature (°F):

Temperature Range	46.6 - 86.0
Mean Temperature	70.4

Size of Treated Fish (in.):

Size Range	1.0 - 9.3

Species Treated:

coho salmon *Oncorhynchus kisutch*

cutthroat trout *O. clarki*

steelhead trout *O. mykiss*

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

muskellunge *Esox masquinongy*

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