Oxytetracycline Medicated Feed Clinical Field Trials - INAD 9006

1999 Annual Summary Report on the Use of Oxytetracycline Medicated Feed in Clinical Field Efficacy Trials

Prepared by:

Bonnie Johnson, Biologist
U.S. Fish and Wildlife Service
Bozeman National INAD Office
Bozeman, Montana

Summary

Oxytetracycline in feed (OTF) was used at four U.S. Fish and Wildlife Service National Fish Hatcheries (NFH) and one tribal hatchery during 1999 to evaluate its efficacy to control mortality caused by bacterial coldwater disease, enteric redmouth, and columnaris in salmonid fish species. The use of OTF has been approved for use in aquaculture by the U.S. Food and Drug Administration (FDA). However, the current label limits drug use to the control of only specific bacterial diseases of fish at water temperatures not below 48.2^{o} F (9^{o} C). Label guidelines do not permit use of oxytetracycline for the control of bacterial coldwater disease, columnaris, enteric redmouth, bacterial kidney disease, or vibriosis. To accommodate the needs of aquaculture and to collect clinical field efficacy data on OTF for the control of these diseases, the FDA has authorized the use of this compound under the Compassionate Investigational New Animal Drug Exemption #9006. In 1999, OTF was administered under INAD #9006 in 127 disease control trials involving approximately 8.5 million fish. Treatment regimes included the use of oxytetracycline medicated feed at 2.5 - 3.5 g/100 lbs fish/day for 10 - 65 days at water temperatures above 48.2^{o} F, and 3.75 - 7 g/100 lbs fish/day for 10 - 17 days at water temperatures below 48.2^{o} F. Approximately 48% of the trials appeared efficacious, 6% appeared ineffective, and 46% were characterized as inconclusive.

Introduction

The current label restricts the use of OTF to the control of furunculosis in salmonids caused by *Aeromonas salmonicida*, and bacterial hemorrhagic septicemia in salmonids and catfish caused by *A. hydrophila* or *Pseudomonas sp.*. Oxytetracycline medicated feed has been shown to be highly effective in controlling these diseases, especially when predisposing environmental stresses are reduced at the time of treatment (Warren 1991). However, the current FDA approved label for OTF limits allowed dosages to a range of 2.5 - 3.75 grams of active drug per 100 pounds of fish per day for 10 days, and limits use to water temperatures "not below 48.2^{o} F (9^{o} C)". These label restrictions severely limit the overall utility of approved OTF use in aquaculture.

Fish culturists have also reported that oxytetracycline medicated feed treatment is a useful tool for the control of bacterial cold water disease (CWD) and columnaris in salmonids. These two diseases, collectively termed "flexibacteriosis" are caused by *Flexibacter psychrophilus* and *F. columnaris*. Enteric redmouth, caused by *Yersinia ruckeri*, vibriosis caused by various members of the genus *Vibrio*, and other less common bacterial diseases of fish also have been found to be responsive to OTF therapy. However, none of these latter uses are yet approved by the FDA.

Purpose

The primary purpose of this report is to summarize the results of calendar year 1999 (CY 99) supplemental OTF field efficacy studies. However, it is also expected that data from these studies will be used to enhance the existing OTF database that has been established from previous years studies for the purpose of expanding and/or extending the approved label for OTF.

Facilities, Materials, and Treatment Procedures

1. Facilities

A total of 4 U.S. Fish and Wildlife Service (FWS) National Fish Hatcheries (NFH) and 1 Tribal Hatchery used OTF to control mortality caused by CWD, enteric redmouth, and columnaris.

2. OTF used in trials

The OTF used in these trials was either Terramycin 100 or Terramycin 100D, both of which contained 100 g active oxytetracycline quaternary salt per pound of premix. All Terramycin 100/100D was supplied by Pfizer, Inc., 1107 South 291 Highway, Lee's Summit, MO. However, oxytetracycline medicated feed was supplied by several different fish feed manufacturers.

3. Drug dosages and duration

As described in the Study Protocol for INAD #9006, Investigators were allowed to use OTF either within the current label range of 2.5 - 3.75 grams of active drug per 100 lbs of fish per day, or at dosages up to 7.0 grams of active drug per 100 lbs of fish per day. Treatment duration was restricted to either 10 days, or "up to 21 days". However, the Wolfcreek NFH deviated from the Study Protocol by treating rainbow trout and brown trout at 2.5 grams of active drug per 100 lbs of fish per day for up to 65 days. Multiple (i.e. repeating) treatments were applied as a result of an extended period of high water temperatures and low dissolved oxygen levels experienced at this facility.

Fish Species and Fish Diseases Involved in CY 99 Trials

1. Species of fish treated

Five salmonid species were treated during CY 99. Fish species treated included: rainbow and steelhead trout (*Oncorhynchus mykiss*); coho salmon (*O. kisutch*); fall chinook salmon (*O. tshawytscha*); and brown trout (*Salmo trutta*).

2. Diseases treated

The diseases treated most frequently during CY 99 were bacterial coldwater disease and columnaris. The other disease treated for was enteric redmouth at the Nez Perce Tribal hatchery.

Data Collected

1. Pathologist's reports

Fish health pathology reports include: 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 a population of test fish. Pathology reports provide essential information if efforts are to expand/extend an existing approved label. Pathologist reports were submitted with studies conducted at Dworshak NFH, Quinault NFH, Willard NFH, and Nez Perce Tribal Hatchery.

2. Mortality data

As stated in the Study Protocol, mortality data was to be collected for at least 10 days prior to treatment, during the treatment period, and for at least 30 days post-treatment. Investigators were strongly encouraged to collect mortality data on a daily basis. However, daily collection of mortality data was not always possible. At production facilities that are understaffed, the collection and enumeration of mortalities can not always be conducted on a daily basis. Therefore, in some cases, mortalities were collected, counted, and recorded only once/twice per week.

Discussion of 1999 Study Results

1. General observations on the efficacy of OTF for the control of bacterial diseases in salmonid and non-salmonid fish

A. Efficacy at 2.5 g/100 lbs fish/day for 13-65 days at water temperatures above 9°C

OTF was used at 2.5 g/100 lbs for 13-65 days in 84 of 127 trials (66%). Trials involved rainbow trout and brown trout at the Wolfcreek NFH diagnosed with columnaris (Table 1-3). Water temperature during treatment was approximately 15.6°C. A total of 34 of these trials appeared efficacious, while 4 trials did not appear efficacious, and 46 trials were characterized as inconclusive. Trials were characterized as inconclusive due to changes that occurred in the fish rearing units (fish populations were split, moved, or increased) during treatment. The Investigator noted that to prevent substantial loss of production fish, OTF is used for the duration of high water temperatures and low oxygen levels (late summer to early fall).

B. Efficacy at 3.5 g/100 lbs fish/day for 10 days at water temperatures above 9°C

OTF was used at 3.5 g/100 lbs for 10 days in 11 of 127 trials (9%). Trials involved fall chinook salmon diagnosed with enteric redmouth at the Nez Perce Tribal hatchery (Table 1). Water temperature during treatment was approximately 10.0°C. In all trials, treatment appeared to be efficacious in controlling mortality.

C. Efficacy at 3.75 g/100 lbs fish/day for 14 days at water temperatures below 9°C

OTF was used at 3.75 g/100 lbs for 14 days in 2 of 127 trials (2%). Both trials involved coho salmon at the Willard NFH diagnosed with bacterial coldwater disease (Table 2.). Water temperature during treatment was approximately 5.3°C. In both trials, treatment did not appear to be efficacious in controlling mortality.

D. Efficacy at 4.0 g/100 lbs fish/day for 10 days at water temperatures below 9°C

OTF was used at 4.0 g/100 lbs for 10 days in 10 of 127 trials (8%). All trials involved coho salmon diagnosed with bacterial coldwater disease at the Quinault NFH (Table 3). Water temperature during treatment was 7.2°C in all trials. Treatment results were inconclusive in all trials.

E. Efficacy at 4.5 g/100 lbs fish/day for 11 days at water temperatures below 9°C

OTF was used at 4.5 g/100 lbs for 11 days in a single trial. The trial involved coho salmon diagnosed with coldwater disease at the Quinalt NFH (Table 1). Water temperature during treatment was 4.4°C. The treatment appeared to be efficacious in controlling mortality.

F. Efficacy at 4.8 g/100lbs fish/day for 11 days at water temperatures below 9°C

OTF was used at 4.8 g/100 lbs for 11 days in a single trial. The trial involved coho salmon diagnosed with coldwater disease at the Quinalt NFH (Table 1). Water temperature during treatment was 8.9°C. The treatment appeared to be efficacious in controlling mortality.

G. Efficacy at 7.0 g/100 lbs fish/day for 13-17 days at water temperatures below 9°C

OTF was used a 7.0 g/100 lbs for 13-17 days in 18 trials (14%). The trials involved rainbow and steelhead trout at the Dworshak NFH (Table1-3). The water temperature during treatment was approximately 8.4°C. A total of 14 of these trials appeared efficacious, while 2 trials did not appear efficacious, and 2 trials were characterized as inconclusive.

Summary of Study Results

Oxytetracycline medicated feed was used at dosages ranging from 2.5 - 7.0 g/100 lbs fish per day. Treatment duration ranged from 10 - 65 days. Five different species of fish were treated with OTF, and trials involved approximately 8.5 million treated fish. Treated fish ranged in size from 1.6-12.2 in. Water temperature during treatment ranged from 4.4-15.6°C, with a mean trial treatment temperature of 13.1°C. Approximately 48% of the trials appeared effective, 6% appeared ineffective, and 46% were characterized as inconclusive. Overall, OTF treatment appeared effective in controlling mortality caused by bacterial coldwater disease, columnaris, or enteric redmouth. Results of trials indicated that mortality decreased during or following the treatment period, and remained at normal levels throughout the post-treatment period. Furthermore, investigators reported no evidence of toxicity or adverse effects related to OTF treatment. However, based on a lack of untreated control fish, replication, randomization, etc., it is understood that these data can only be considered as ancillary data. None-the-less, the ancillary data described above should provide useful corroborative data to support a future expanded label claim for OTF. It is anticipated that additional ancillary efficacy data will continue to be collected under INAD #9006. In future trials conducted under INAD 9006, efforts will be directed towards the generation of higher quality data.

References

Warren, J.W. 1991. Diseases of hatchery fish. U.S. Fish and Wildlife Service, Portland, Oregon, 92 p.

Table 1. Summary of 1999 Oxytetracycline Medicated Feed Efficacy Results - Efficacious Studies

Hatchery	Number of Efficacious Trials	Fish Size (in.)	Fish Species	Number of Fish	Disease	Number of Treatment Days	Dose (g/100 lbs)	Temp. (ºC)
Wolf Creek NFH	3	7.00	RBT	27,650	Columnaris	13-14	2.50	15.6
	6	7.00	RBT	77,000	Columnaris	21	2.50	15.6
	2	7.00	RBT	17,100	Columnaris	31-38	2.50	15.6
	4	7.00	RBT	132,250	Columnaris	48-52	2.50	15.6
	19	7.00	RBT	258,550	Columnaris	65	2.50	15.6
Nez Perce Tribal Hatchery	11	3.22	FCS	350,999	ERM	10	3.50	10.0
Quinalt NFH	1	1.59	COS	342,000	CWD	11	4.50	4.4
Quinalt NFH	1	4.75	COS	70,000	CWD	11	4.80	8.9
Dworshak NFH	1	12.20	RBT	4,700	CWD	13	7.0	6.3
	8	4.1-4.7	STT	243,730	CWD	14	7.0	9.0
	5	5.0-5.4	STT	141,577	CWD	15	7.0	8.3

Table 2. Summary of 1999 Oxytetracycline Medicated Feed Efficacy Results - <u>Non-efficacious Studies</u>

Hatchery	# of Non-efficacious Trials	Fish Size (in.)	Fish Species	Number of Fish	Disease	Number of Treatment Days	Dose (g/100 lbs)	Temp. (ºC)
Wolf Creek NFH	1	7.00	RBT	18,225	Columnaris	44	2.50	15.6
	3	7.00	RBT	25,975	Columnaris	65	2.50	15.6
Willard NFH	2	1.8-2.5	COS	5,422,079	CWD	14	3.75	5.0-5.6
Dworshak NFH	1	5.31	STT	27,000	CWD	14	7.0	8.6
	1	11.9	RBT	4,748	CWD	17	7.0	5.4

Table 3. Summary of 1999 Oxytetracycline Medicated Feed Efficacy Results - <u>Inconclusive Studies</u>

Hatchery	Number of Inconclusive Trials	Fish Size (in.)	Fish Species	Number of Fish	Disease	Number of Treatment Days	Dose (g/100 lbs)	Temp. (ºC)
Wolf Creek NFH	13	7.0	RBT	174,665	Columnaris	13-17	2.50	15.6
	11	7.0	RBT	150,250	Columnaris	21	2.50	15.6
	2	7.0	RBT	33,525	Columnaris	28-31	2.50	15.6
	8	7.0	RBT	229,175	Columnaris	44	2.50	15.6
	11	7.0	RBT	126,525	Columnaris	52	2.50	15.6
	1	7.0	BNT	53,500	Columnaris	55	2.50	15.6
Quinalt NFH	10	5.0	COS	605,000	CWD	10	4.0	7.2
Dworshak NFH	2	3.8-4.7	STT	52,324	CWD	14-15	7.0	8.8-9.0

Table 4. Summary Data Regarding 1999 Oxytetracycline Medicated Feed Efficacy Studies

Total Number of Fish Treated:	**8,588,547**
Number of fish treated in efficacious studies	1,665,556
Number of fish treated in non-efficacious studies	5,498,027
Number of fish treated in inconclusive studies	1,424,964
Total Number of Rearing Units Treated:	**127**
Rearing Units in Efficacious Studies	61
Rearing Units in Non-efficacious Studies	8
Rearing Units in Inconclusive Studies	58

Treatment Regimes Used:

2.5 g/100 lbs fish/day for 13-65 days (above 9°C)	84 trials
3.5 g/100 lbs fish/day for 10 days (above 9°C)	11 trials
3.75 g/100 lbs fish/day for 14 days (below 9°C)	2 trials
4.0 g/100 lbs fish/day for 10 days (below 9°C)	10 trials
4.5 g/100 lbs fish/day for 11 days (below 9°C)	1 trial
4.8 g/100 lbs fish/day for 11 days (below 9°C)	1 trial
7.0 g/100 lbs fish/day for 13-17 days (below 9°C)	18 trials

Treatment Water Temperature (°C):

Temperature Range	4.4 - 15.6
Mean Trial Temperature	13.1

Size of Treated Fish (in.):

Size Range	1.6 - 12.2

Species Treated: rainbow trout (*Oncorhynchus mykiss*)
fall chinook salmon (*O. tshawytscha*)
coho salmon (*O. kisutch*)
brown trout (*Salmo trutta*)
steelhead trout (*O. mykiss*)