Oxytetracycline Medicated Feed Clinical Field Trials - INAD 9332

1998 Annual Summary Report on the Use of Oxytetracycline Medicated Feed in Field Efficacy Trials

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Summary

Oxytetracycline as a feed additive (OTF) was used at 19 state fish hatcheries, 2 tribal hatcheries, and 4 private hatcheries during 1998 to evaluate its efficacy to control mortality caused by bacterial coldwater disease, columnaris, flexibacteriosis, systemic flexibacteriosis, and vibriosis in salmonids. 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 specific fish species at water temperatures not below 48.2° F (9° C). Label guidelines do not permit the 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 pivotal and ancillary clinical field 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 (INAD) Exemption #9332. In 1998, OTF was administered under INAD #9332 in 63 disease control/prevention trials and involved approximately 26.7 million fish. Standard treatment regimes included the use of OTF at 2.5 - 3.75 g/100 lbs fish/day for 10 days; and 10.0 g/100 lbs fish/day for 14 days. In a few select trials fish were fed oxytetracycline medicated feed at 1.9 - 2.3 g/100 lbs fish/day for 15 days, 3.86-9.2 g/100 lbs fish/day for 11 - 15 days, and 10.1 - 17.0 g/100 lbs fish/day for 14 - 15 days. Approximately 86% of trials appeared efficacious, 8% appeared ineffective, and 6% were characterized as inconclusive.

Introduction

The current label for OTF use in aquaculture limits use to the control of furunculosis in salmonids caused by *Aeromonas salmonicida*, and the control of 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° F (9° C)". These label restrictions severely limit the overall utility of approved OTF use in aquaculture.

Fish culturists have also reported that OTF 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 approved by the FDA.

Purpose of Report

The primary purpose of this report is to summarize the results of calendar year 1998 (CY 98) supplemental OTF field efficacy studies. However, it is also expected 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 19 state fish hatcheries, 2 tribal hatcheries, and 4 private hatcheries (25 total facilities) used OTF to control mortality caused by CWD, columnaris, flexibacteriosis, systemic flexibacteriosis, and vibriosis.

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 #9332, 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 for 10 days (~32% of studies), or at 10.0 grams of active drug per 100 lbs of fish per day for 14 days (~41% of studies). However, a number of trials (~27%) deviated from the protocol during CY 98. In these trials, fish were fed at rates of 1.9 - 17.0 grams of active drug per drug/100 lbs fish/day for periods of time ranging from 11 - 15 days.

Fish Species and Fish Diseases Involved in 1998 Trials

1. Species of fish treated

Five salmonid species were treated during CY 98. Species treated included rainbow and steelhead trout (*Oncorhynchus mykiss*); coho salmon (*O. kisutch*); chinook salmon (*O. tshawytscha*); and cutthroat trout (*O. clarki*).

2. Diseases treated

The diseases treated most frequently during CY 98 were bacterial coldwater disease and flexibacteriosis. Other diseases treated were columnaris, systemic flexibacteriosis, and vibriosis.

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. Approximately 40% of CY 98 trials included pathologist’s reports.

2. Mortality data

As stated in the Study Protocol, mortality data was to be collected 5 days prior to treatment, during the treatment period, and for at least 20 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 Study Results:

General observations on the efficacy of OTF for the control of bacterial diseases of salmonids (Note: A summary of all OTF studies conducted during CY 98 under INAD #9332 is presented in Table 1.)

A. Efficacy at 1.9 - 2.3 g/100 lbs fish/day for 15 days at water temperatures above 48.2° F

OTF was used at 1.9 - 2.3 g/100 lbs of fish for 15 days in 2 trials (Table 2). Both trials appeared efficacious in controlling mortality caused by flexibacteriosis in cutthroat trout.

B. Efficacy at 2.5 - 3.75 g/100 lbs fish/day for 10 days at water temperatures below 48.2° F

OTF was used at 2.5 - 3.75 g/100 lbs of fish for 10 days in 6 trials (Table 2). All of these trials appeared efficacious in controlling mortality caused by CWD and systemic flexibacteriosis in rainbow trout and coho salmon.

C. Efficacy at 2.5 - 3.75 g/100 lbs fish/day for 10 - 15 days at water temperatures above 48.2° F

OTF was used at 2.5 - 3.75 g/100 lbs of fish for 10 - 15 days in 15 trials (Tables 2 - 4). OTF treatment appeared efficacious in 13 trials in controlling mortality caused by CWD, columnaris, flexibacteriosis, and systemic flexibacteriosis. Fish species treated included cutthroat trout, rainbow trout, steelhead trout, and chinook salmon. OTF treatment did not appear efficacious in 1 trial, and treatment results were characterized as inconclusive in 1 trial.

D. Efficacy at 3.86 - 9.2 g/100 lbs fish/day for 11 days at water temperatures below 48.2° F

OTF was used at 3.86 - 9.2 g/100 lbs of fish for 11 days in 1 trial (Table 2). This trial appeared efficacious in controlling mortality caused by CWD in coho salmon.

E. Efficacy at 3.86 - 9.2 g/100 lbs fish/day for 10 - 15 days at water temperatures above 48.2° F

OTF was used at 3.86 - 9.2 g/100 lbs of fish for 10 - 15 days in 10 trials (Tables 2 - 4). OTF treatment a appeared efficacious in 6 trials in controlling mortality caused by CWD, flexibacteriosis, and systemic flexibacteriosis. Fish species treated included cutthroat trout, rainbow trout, and steelhead trout.

OTF treatment did not appear efficacious in 2 trials, and treatment results were characterized as inconclusive in 2 trials.

F. Efficacy at 10.0g/100lbs fish/day for 10 days (3 times) at water temperatures below 48.2°F

OTF was used at 10.0 g/100lbs fish/day for 10 days in 1 trial (Table 2.). This trial involving coho salmon with CWD held at South Fork Klaskanine Hatchery. Treatment deviated from the Study Protocol as fish were treated with three repetitive 10 day treatments of OTF (each treatment separated by a 2 day period of regular, unmedicated feed). Percent daily mortality dropped sharply after the first 10 day treatment and continued to decrease throughout the remaining treatment days. OTF treatment appeared efficacious.

G. Efficacy at 10.0 g/100 lbs fish/day for 11 - 15 days at water temperatures below 48.2° F

OTF was used at 10.0 g/100 lbs of fish for 11 - 15 days in 4 trials (Tables 2 & 3). OTF treatment appeared efficacious in 3 trials in controlling mortality caused by CWD in coho salmon and steelhead trout. OTF treatment did not appear efficacious in 1 trial.

H. Efficacy at 10.0 g/100 lbs fish/day for 10 - 15 days at water temperatures above 48.2° F

OTF was used at 10.0 g/100 lbs of fish for 10 - 15 days in 21 trials (Tables 2 & 3). OTF treatment appeared efficacious in 20 trials in controlling mortality caused by CWD, flexibacteriosis, and columnaris. Fish species treated included cutthroat trout, rainbow trout, steelhead trout, coho salmon, and chinook salmon. OTF treatment did not appear efficacious in 1 trial.

I. Efficacy at 10.1 - 17.0 g/100 lbs fish/day for 14 days at water temperatures below 48.2° F

OTF was used at 10.1 - 17.0 g/100 lbs of fish for 14 days in 1 trial (Table 4). Results of OTF treatment to control mortality caused by CWD in coho salmon in this trial were characterized as inconclusive.

J. Efficacy at 10.1 - 17.0 g/100 lbs fish/day for 15 days at water temperatures above 48.2° F

OTF was used at 10.1 - 17.0 g/100 lbs of fish for 15 days in 2 trials (Table 2). OTF treatment in both trials appeared efficacious in controlling mortality caused by CWD and flexibacteriosis in rainbow trout and cutthroat trout, respectively.

Summary of Study Results

Oxytetracycline medicated feed was used at dosages ranging from 1.9 - 17.0 g/100lbs fish per day. Treatment duration ranged from 10 - 15 days. Five different species of fish were treated with OTF, and trials involved approximately 26.7 million fish. Treated fish ranged in size from 0.75 - 8.0 in. Water temperature during treatment ranged from 37.0 - 62.5 °F, with a mean treatment temperature of 51.6 °F. Approximately 86% of trials appeared efficacious, 8% appeared ineffective, and 6% were characterized as inconclusive. Five trials involved the use of control fish and ~40% of trials included pathologist's reports. Overall, OTF appeared effective in controlling mortality caused by bacterial coldwater disease or flexibacteriosis. 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 general 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 #9332. In future trials conducted under INAD #9332, 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. This table is generated form database on Bonnie’s computer (pages).

Table 2. Summary of 1998 Oxytetracycline Medicated Feed Efficacy Results - Efficacious Studies

Hatchery	Number of Trials	Fish Species	Fish Size (inches)	Number of Fish	Disease	Number of Treatment Days	Dose (g/100 lbs)	Temp. (°F)
Washoe Park Trout Hatchery	2	CUT	0.75 - 1.00	57,000	Flexibacteriosis	15	1.9 - 2.3	56
Hidden Falls Hatchery	5	COS	4.40 - 4.60	1,595,268	CWD	10	2.5 - 3.75	43.0 - 44.5
Washoe Park Trout Hatchery	2	CUT	0.75 - 1.00	43,000	Flexibacteriosis	15	2.5 - 3.75	56
Murray Springs Trout Hatchery	7	CUT	1.00	505,000	Flexibacteriosis	10 - 11	2.5 - 3.75	52
Clark Fork Hatchery	2	RBT	3.90 & 6.30	381,404 125,714	Systemic Flexibacteriosis	10	2.5 - 3.75	49.5 42.0
Grace Fish Hatchery	1	RBT	3.20	145,800	Systemic Flexibacteriosis	10	2.5 - 3.75	52
Cowlitz Salmon Hatchery	1	SCS	6.60	280,000	CWD	10	2.5 - 3.75	53
Clearwater Hatchery	1	STT	3.80	620,000	Columnaris	10	2.5 - 3.75	57
Sheep Creek Hatchery	1	COS	3.80	250,692	CWD	11	3.8 - 9.2	37
Washoe Park Trout Hatchery	2	CUT	0.75 - 1.50	194,500	Flexibacteriosis	14 - 15	3.8 - 9.2	56

Table 2. Summary of 1998 Oxytetracycline Medicated Feed Efficacy Results - Efficacious Studies (cont.)

Hatchery	Number of Trials	Fish Species	Fish Size (inches)	Number of Fish	Disease	Number of Treatment Days	Dose (g/100 lbs)	Temp. (°F)
American River Hatchery	2	RBT	2.20 - 2.40	1,226,000	CWD	10	3.8 - 9.2	54 - 55
Grace Fish Hatchery	1	RBT	3.90	67,900	Systemic Flexibacteriosis	10	3.8 - 9.2	52
Warm Springs Salmon and Trout Hatchery	1	STT	2.10	23,104	Flexibacteriosis	10	3.8 - 9.2	54
Big Creek Hatchery	1	COS	3.00	600,000	CWD	14	10	49.5
Cascade Hatchery	1	COS	1.80	6,513,988	CWD	14	10	46
Oxbow Hatchery	1	COS	3.30	646,000	CWD	15	10	42.6
South Fork Klaskanine Hatchery	1	COS	2.00	618,203	CDW	30[1]	10	48.1
Murray Springs Trout Hatchery	3	CUT	1.00	254,000	Flexibacteriosis	10 - 11	10	52
Clearwater Hatchery	1	RBT	8.00	32,000	Columnaris	14	10	57
Leaburg Hatchrey	1	RBT	3.60	749,399	CWD	12	10	56
Cowlitz Salmon Hatchery	7	SCS	4.5 - 5.0	758,705	CWD	13 - 14	10	53

Table 2. Summary of 1998 Oxytetracycline Medicated Feed Efficacy Results - Efficacious Studies (cont.)

Hatchery	Number of Trials	Fish Species	Fish Size (inches)	Number of Fish	Disease	Number of Treatment Days	Dose (g/100 lbs)	Temp. (°F)
Dexter Ponds	1	SCS	5.20	221,000	Columnaris	14	10	60
Bonneville Hatchery	2	STT	1.7 - 2.0	779,965	CWD	14	10	49
Cowlitz Trout Hatchery	1	STT	3.80	1,206,000	CWD	14	10	50
Mad River Hatchery	1	STT	2.00	431,300	Flexibacteriosis	14	10	52
South Santiam Hatchery	1	STT	3.20	234,063	CWD	15	10	52.5
Willamette Hatchery	2	STT	2.1 - 4.7	351,522	CWD	14	10	48 - 54
Washoe Park Trout Hatchery	1	CUT	0.75	10,000	Flexibacteriosis	15	10.1 - 17.0	56
Big Springs Trout Hatchery	1	RBT	2.70	25,230	CWD	15	10.1 - 17.0	55

[1] Fish were given 3 repetitive 10 day treatments (2 days on unmedicated feed between each 10 day treatment period)

Table 3. Summary of 1998 Oxytetracycline Medicated Feed Efficacy Results - Non-efficacious Studies

Hatchery	Number of Trials	Fish Species	Fish Size (inches)	Number of Fish	Disease	Number of Treatment Days	Dose (g/100 lbs)	Temp. (°F)
Grace Fish Hatchery	1	RBT	4.0	51,000	Flexibacteriosis	10	2.5 - 3.75	52
Salmon River Tribal Hatchery	1	COS	1.90	740,405	CWD	10	3.8 - 9.2	54
Washoe Park Trout Hatchery	1	CUT	12.10	2,200	Flexibacteriosis	15	3.8 - 9.2	50
Cascade Hatchery	1	COS	2.1	4,430,112	CWD	14	10	46.1
Willamette Hatchery	1	RBT	1.25	351,474	CWD	14	10	54

Table 4. Summary of 1998 Oxytetracycline Medicated Feed Efficacy Results - <u>Inconclusive Studies</u>

Hatchery	Number of Trials	Fish Species	Fish Size (inches)	Number of Fish	Disease	Number of treatment days	Dose (g/100 lbs)	Temp. (°F)
Central Coast Salmon Enhan.	1	CKS	5.7	35,420	Vibriosis	10	2.5 - 3.75	62.5
Lower Elwha Tribal Hatchery	1	COS	2.5	653,903	CWD	14	3.8 - 9.2	48.5
American River Hatchery	1	RBT	1.5	865,000	CWD	10	3.8 - 9.2	54
Lower Elwha Tribal Hatchery	1	COS	3.4	632,393	CWD	14	10.1 - 17.0	48

Table 5. Summary Data Regarding 1998 Oxytetracycline Medicated Feed Efficacy Studies

Total Fish Treated:	**26,708,664**
Number of fish treated in efficacious studies	18,946,757
Number of fish treated in non-efficacious studies	5,575,191
Number of fish treated in inconclusive studies	2,186,716
Total number of studies:	**63**
Efficacious studies	54
Non-efficacious studies	5
Inconclusive studies	4

Studies that Included Control Fish:

Study Number: 9332-98-020; 9332-98-024; 9332-98-028; 9332-98-035; and 9332-98-050

Treatment Regimes Used:

1.9 - 2.3 g/100 lbs fish/day for 15 days (above 48.2°F)	2 trials
2.5 - 3.75 g/100 lbs fish/day for 10 days (below 48.2°F)	6 trials
2.5 - 3.75 g/100 lbs fish/day for 10 - 15 days (above 48.2°F)	15 trials
3.8 - 9.2 g/100 lbs fish/day for 11 days (below 48.2°F)	1 trial
3.8 - 9.2 g/100 lbs fish/day for 10 - 15 days (above 48.2°F)	10 trials
10 g/100 g/100 lbs fish/day for 10 days (below 48.2°F)	1 trial
10 g/100 g/100 lbs fish/day for 10 - 15 days (above 48.2°F)	21 trials
10 g/100 g/100 lbs fish/day for 11 - 15 days (below 48.2°F)	4 trials
10.1 - 17.0 g/100 lbs fish/day for 14 days (below 48.2°F)	1 trial
10.1 - 17.0 g/100 lbs fish/day for 15 days (above 48.2°F)	2 trials

Treatment Water Temperature (°F):

Temperature Range	37.0 - 62.5
Mean Temperature	51.6

Size of Treated Fish (in.):

Size Range	0.75 - 8.0

Species Treated: rainbow and steelhead trout (*Oncorhynchus mykiss*)
coho salmon (*O. kisutch*)
chinook salmon (*O. tshawytscha*)
cutthroat trout (*O. clarki*)