

Oxytetracycline Medicated Feed Clinical Field Trials - INAD 9332

Year 2002 Annual Summary Report on the Use of Oxytetracycline Medicated Feed as a Marking Agent in Field Efficacy Trials

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Summary

Oxytetracycline medicated feed (OTF-M) has been shown to be an effective and convenient marking agent for use on early life stages of fish. Large numbers of young fish can be marked simultaneously by feeding fish a standard dosage of OTF-M for up to 10 d. In many cases, OTF-M is the only viable option, other than immersion marking with water-soluble oxytetracycline, for permanently marking large numbers of small fish for the purpose of evaluating fishery management strategies. Oxytetracycline medicated feed has been approved for use in aquaculture for limited therapeutic uses and for marking skeletal tissue in Pacific salmon in the United States by the U.S. Food and Drug Administration (FDA). To accommodate the needs of aquaculture and to collect pivotal and ancillary clinical field data on OTF-M for the marking skeletal tissue in a variety of salmonid species, the FDA has authorized the use of this compound under the Compassionate Investigational New Animal Drug (INAD) Exemption #9332. A number of trials were conducted in calendar year 2002 under INAD #9332 to evaluate

the efficacy of OTF-M as a marking agent of skeletal tissue in a variety of salmonids. Twenty-five such trials that involved approximately 4.8 million fish were conducted at seven state fish hatcheries during this period. Efficacy was determined by whether or not a “readable” mark could be observed on skeletal tissue of treated fish. Standard treatment regimens included the use of OTF-M at 2.5 - 3.76 g/100 lbs fish/d for 10 - 14 d; and 10.0 g/100 lbs fish/d for 8 - 14 d. In a few select trials fish were fed OTF-M at 3.9 - 4.2 g/100 lbs fish/d for 10 - 12 d. All trials appeared efficacious.

Introduction

The current label for OTF-M use in aquaculture limits its use to marking of skeletal tissue in Pacific salmonids only. The current U. S. Food and Drug Administration (FDA) approved label for OTF-M limits the allowed dosages to 250 mg/kg of fish/d (i.e., 11.4 g OTF-M/100 lbs fish/d) for 4 d in salmon less than 30 gm with a 7 d withdrawal time. These label restrictions severely limit the overall utility of approved OTF-M use in aquaculture.

Fish culturists have reported that OTF-M treatment is a useful tool for marking the skeletal tissue in rainbow trout, cutthroat trout, and kokanee salmon that are > 2 gms, which is the largest fish size that can be treated with soluble oxytetracycline in a bath. Marks were visible immediately after the treatment period, and they were still visible for several months afterwards when skeletal tissue of fish were evaluated. Side by side comparisons have been conducted using different drug dosages of OTF-M to mark

skeletal tissue of test fish with the same efficacious results, and without any toxic or adverse effects to the fish. However, using OTF-M on non-pacific salmonids for marking skeletal tissue has not yet been approved by the FDA.

Purpose of Report

The primary purpose of this report was to summarize the results of calendar year 2002 (CY 02) supplemental OTF-M field efficacy studies. However, it is also expected data from these studies will be used to enhance the existing OTF-M database that has been established from previous years studies for the purpose of expanding and/or extending the approved label for OTF-M.

Facilities, Materials, and Treatment Procedures

1. Participating Facilities

A total of seven state fish hatcheries used OTF-M for marking skeletal tissue of test fish.

2. OTF-M used in efficacy trials

The OTF-M used in CY 02 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. All OTF-M was prepared with Pfizer brand product, and was prepared by one of several commercial fish feed manufacturers.

3. Drug dosages and duration

As described in the Study Protocol for INAD #9332, Investigators were allowed to use OTF-M either within the current label range of 2.5 - 3.76 gm of active drug per 100 lbs of fish/d for 10 - 14 d (~48% of studies), or at 10.0 gm of active drug per 100 lbs of fish/d for 8 - 14 d (~40% of studies). Investigators deviated from the protocol in few trials (~12%) when fish were fed at rates ranging from 3.91 to 4.20 gm of active drug per drug/100 lbs fish/d for periods of time ranging from 10 to 12 ds.

Fish Species Involved in CY 02 Efficacy Trials

1. Species of fish treated

The following three salmonid species were marked during CY 02:

rainbow trout *Oncorhynchus mykiss*

cutthroat trout *O. clarki*

kokanee salmon *O. nerka*

2. Marking

Fish were treated with OTF-M to provide a readable mark on skeletal tissue that could be used as an important fishery management tool.

Data Collected

1. Pathologist's reports

No pathology reports were submitted during CY 02 studies.

2. Efficacy of marking procedure

Samples of treated fish were collected, processed, and evaluated for a mark on skeletal tissue using standard procedures.

Discussion of Study Results:

- 1. General observations on the efficacy of OTF-M for marking of skeletal tissue of salmonids** - Efficacy was determined by whether or not a “readable” mark could be observed on skeletal tissue of treated fish. (Note: A summary of all OTF-M studies conducted during CY 02 under INAD #9332 is presented in Table 3.)

A. Efficacy at 3.75 g/100 lbs fish/d for 10 d at water temperatures below 48.2° F

OTF-M was used at 3.75 g/100 lbs of fish/d for 10 d in two trials involving rainbow trout (Table 1). OTF-M treatment appeared efficacious in both.

B. Efficacy at 2.50 - 3.76 g/100 lbs fish/d for 10 - 14 d at water temperatures above 48.2° F

OTF-M was used at 2.50 - 3.76 g/100 lbs of fish/d for 10 - 14 d in 10 trials involving rainbow trout, cutthroat trout, and kokanee salmon (Table 1).

OTF-M treatment appeared efficacious in all trials with.

C. Efficacy at 3.91 - 4.20 g/100 lbs fish/d for 10 - 12 d at water temperatures above 48.2° F

OTF-M was used at 3.91 - 4.20 g/100 lbs of fish/d for 10 - 12 d in three trials involving kokanee salmon (Table 1). OTF-M treatment appeared efficacious in all trials.

D. Efficacy at 10g/100lbs fish/d for 8 - 14 d at water temperatures above 48.2°F

OTF-M was used at 10 g/100lbs fish/d for 8 - 14 d in 10 trials involving rainbow trout, cutthroat trout, and kokanee salmon (Table 1). OTF-M treatment appeared efficacious in all trials.

2. Observed Toxicity

No toxicity or adverse effects relating to OTF-M treatments were reported in any of the trials conducted in CY 02. In one trial, the Investigator noted that mortality was higher than normal during the post-treatment period; however, the elevated mortality was probably caused by overcrowded culture conditions.

Summary of Study Results

Oxytetracycline medicated feed was used to mark skeletal tissue of test fish at dosages ranging from 2.50 to 10.0 g/100lbs fish/d for durations ranging from 8 to 14 d. Three different species of fish were treated with OTF-M, and trials involved approximately 4.8 million fish. Treated fish ranged in size from 1.7 - 6.0 in. Overall mean water temperature during treatments was 52.6 °F (range, 41.5 - 58.0 °F). All treatment trials appeared efficacious. One trial involved the use of control fish. None of the trials included a pathologist's reports documenting health of test fish prior to or after treatment. Overall, OTF-M appeared effective in creating a readable mark on the

skeletal tissues of the treated fish. Furthermore, Investigators reported no evidence of toxicity or adverse effects related to OTF-M treatment. Post-treatment mortality in one trial was higher than normal, but was probably due to poor culture conditions (i.e., overcrowding in the rearing units). 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 in this report should provide useful corroborative data to support a future expanded label claim for OTF-M. It is anticipated that additional ancillary skeletal tissue marking efficacy data will continue to be collected under INAD #9332. In future trials conducted under INAD #9332 for the purpose of marking fish, efforts will be directed towards the generation of higher quality data.

Table 1. Summary of Year 2002 Oxytetracycline Medicated Feed Efficacy Results - Efficacious Studies

Hatchery	Number of Trials	Fish Species	Fish Size (inches)	Fish Number	Use of Feed	Dose (g/100 lbs)	Treatment Duration (days)	Temp. (°F)
Big Springs Trout SFH	1	CUT	5.90	10,000	Marking	2.50 - 3.75	10	51.0
	1	KOE	2.75	207,000	Marking	2.50 - 3.75	10	55.0
	8	RBT	2.9 - 6.0	922,610	Marking	2.50 - 3.76	10 - 14	51 - 55.5
Jocko River Trout SFH	1	RBT	3.60	45,425	Marking	2.50 - 3.75	10	41.5
Washoe Park Trout SFH	1	RBT	2.70	114,115	Marking	2.50 - 3.75	10	45
Big Springs Trout SFH	2	KOE	2.90	173,712	Marking	3.91 - 4.20	10	55.0
Bluewater Springs Trout SFH	1	KOE	4.85	50,000	Marking	3.91 - 4.20	12	58.0
Yellowstone R. Trout SFH	1	CUT	2.30	30,000	Marking	10.0	14	52.0
Bluewater Springs Trout SFH	1	KOE	2.80	100,000	Marking	10.0	10	58.0
Roaring Judy SFH	4	KOE	1.70	2,814,866	Marking	10.0	8	51.0
Bluewater Springs Trout SFH	2	RBT	2.7 - 2.9	100,000	Marking	10.0	10 - 13	58.0
Giant Springs Trout SFH	2	RBT	2.6 - 2.9	230,000	Marking	10.0	14	54.0

**Table 2. Summary Data Regarding Year 2002 Oxytetracycline
Medicated Feed Efficacy Studies**

Total Number of Fish Treated: **4,797,728**

Number of fish in efficacious studies 4,797,728

Total number of studies: **25**

Number of Efficacious studies 25

Treatment Regimens Used:

3.75 g/100 lbs fish/day for 10 days (below 48.2°F)	2 trials
2.5 - 3.76 g/100 lbs fish/day for 10 - 14 days (above 48.2°F)	10 trials
3.91 - 4.20 g/100 lbs fish/day for 10 - 12 days (above 48.2°F)	3 trials
10.0 g/100 lbs fish/day for 8 - 14 days (above 48.2°F)	10 trials

Treatment Water Temperature (°F):

Temperature Range	41.5 - 58.0
Mean Temperature	52.6

