



INAD 009006 G 0103

Dave Erdahl, Ph.D.
National INAD Coordinator
USFWS Fish Technology Center
4050 Bridger Canyon Road
Bozeman, Montana 59715

DEC 31 2003

Dear Dr. Erdahl:

We refer to your submission dated July 29, 2003, to your investigational new animal drug (INAD) file for the use of oxytetracycline-medicated feed to control mortality caused by bacterial diseases in various species of fish. You requested review of the effectiveness section of a Freedom of Information (FOI) Summary. The proposed claims for the use of oxytetracycline-medicated feed administered at 3.75 g/100 pounds of fish/day for 10 consecutive days are for the control of mortality due to coldwater disease caused by *Flavobacterium columnare* in freshwater-reared salmonids and for the control of mortality due to columnaris disease caused by *Flavobacterium columnare* in freshwater-reared steelhead trout.

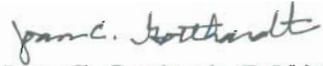
We have reviewed your submission and have the following comments.

We have revised the FOI Summary you submitted. A copy of the FOI Summary is included with this letter. The safety and effectiveness sections of the FOI Summary for your proposed claims are complete. The general information section can be completed along with the other sections of the FOI Summary at the time an approval is requested.

Future correspondence regarding this submission should be identified by the date of this letter and our file number, INAD 009006 G 0103, and be submitted to the Document Control Unit, HFV-199. Please include only one request per submission, clearly stating the request in the first paragraph of the submission.

If you have any questions regarding this letter, please contact Dr. Donald Prater, Leader, Aquaculture Drugs Team, at 301-827-7567.

Sincerely yours,



Joan C. Gotthardt, D.V.M.
Director, Division of Therapeutic
Drugs for Food Animals
Office of New Animal Drug Evaluation
Center for Veterinary Medicine

Enclosure

Date of Approval: _____

FREEDOM OF INFORMATION SUMMARY

NADA xxx-xxx

(drug's proprietary name)

oxytetracycline (monoalkyl trimethyl ammonium salt)

“For the control of mortality in freshwater-reared salmonids caused by coldwater disease and for the control of mortality in freshwater-reared steelhead trout caused by columnaris disease.”

Sponsored by:

(sponsor's name)

1. GENERAL INFORMATION

a. File Number:	NADA xxx-xxx
b. Sponsor:	To be determined
	Drug Labeler Code: (from 21 CFR 510.600)
c. Established Name:	Oxytetracycline (monoalkyl trimethyl ammonium salt)
d. Proprietary Name:	To be determined
e. Dosage Form:	Type A medicated article
f. How Supplied:	To be determined
g. How Dispensed:	Over the counter
h. Amount of Active Ingredient:	10 or 50 grams per pound of powder
i. Route of administration:	Oral via feed
j. Species/Class:	Fresh-water reared salmonids Fresh-water reared steelhead trout
k. Recommended Dosage:	3.75 g oxytetracycline/100 lbs of fish/day for 10 consecutive days
l. Pharmacological Category:	Antimicrobial
m. Indications:	For the control of mortality due to coldwater disease caused by <i>Flavobacterium psychrophilum</i> . For the control of mortality in freshwater-reared steelhead trout <i>Oncorhynchus mykiss</i> due to columnaris disease caused by <i>Flavobacterium columnare</i>
n. Effect of the Supplement	Addition of two disease claims with point doses for a previously approved species.

2. EFFECTIVENESS

a. Dosage Characterization

The dose selected for use in the studies demonstrating substantial evidence of effectiveness was selected based on the approved dose range of 2.5-3.75 g of oxytetracycline per 100 pounds of fish per day and the approved duration of 10 consecutive days.

b. Substantial Evidence

1) Study Number BOZ-98-OTF Study #1

Title: “The Efficacy of Oxytetracycline-Medicated Feed to Control Mortality of Juvenile Steelhead Trout *Oncorhynchus mykiss* Caused by Columnaris, (Causative Agent *Flavobacterium columnare*)”

Study Director: James D. Bowker

Study Location: US Fish and Wildlife Service
Coleman National Fish Hatchery
Anderson, CA

General Design of the Study:

- a. Purpose: To evaluate the effectiveness of oxytetracycline administered in feed at a dose of 3.75 g oxytetracycline/100 pounds of fish/day for 10 consecutive days to control mortality associated with columnaris disease caused by *F. columnare* in steelhead trout.
- b. Animals: Approximately 17,320 steelhead trout juveniles.
- c. Test article/controls: The test article was a Type C medicated feed containing oxytetracycline that delivered a dose of 3.75 g oxytetracycline/100 pounds of fish/day for 10 consecutive days. The control was a non-medicated commercial salmonid feed.
- d. Study Design: Fish were examined (body surface, fins, gills, and internal organs) and gill, kidney, spleen, and external skin lesion inocula were cultured and evaluated for the presence of systemic bacteria. Columnaris disease was confirmed as the cause of increased mortality in two tanks serving as the source of test fish. Test fish were systematically transferred to six test tanks. Untreated control and oxytetracycline-medicated groups were tested in triplicate. Fish were observed and mortality recorded for four days prior to treatment. Fish were treated with medicated feed (3.75 g oxytetracycline/100 pounds of fish/day) for 10 consecutive days. Fish were observed and mortality recorded for an additional 10 days. The concentration of oxytetracycline in the medicated feed was assayed to verify the dosage.

- e. **Parameters Measured:** Mortality and water quality parameters were recorded.

Statistical Analysis: The percent cumulative mortality rates for the untreated control and oxytetracycline-medicated control groups were compared using the GENMOD procedures in SAS, with a binomial error distribution and a logit link function. Overdispersion was allowed for using the PSCALE option and included the fixed effects of treatment group and the natural logarithm of pre-treatment mortality rates as a covariate.

Results: Mortality rates are included in the following table.

Table 1. Mortality rates for the 10-day treatment period and 10-day post-treatment period for a field study involving a columnaris infection in steelhead trout.

Treatment Group	Mean Cumulative Mortality	Mean Percent Cumulative Mortality
Control	673	28.8
Oxytetracycline-treated	309	13.4
p-value		<0.0001

Conclusions: The results demonstrate the effectiveness of oxytetracycline-medicated feed administered at 3.75 g oxytetracycline/100 pounds of fish/day for 10 consecutive days for the control of mortality associated with columnaris disease, caused by *F. columnare*, in steelhead trout reared in freshwater.

2) Study Number BOZ-98-OTF-02

Title: “The Efficacy of Oxytetracycline-Medicated Feed to Control Mortality Caused by Columnaris, Causative Agent *Flavobacterium columnare*, of Juvenile Steelhead Trout *Oncorhynchus mykiss*”

Study Director: James D. Bowker

Study Location: US Fish and Wildlife Service
Coleman National Fish Hatchery
Anderson, CA

General Design of the Study:

- Purpose:** To evaluate the effectiveness of oxytetracycline administered in feed at a dose of 3.75 g oxytetracycline/100 pounds of fish/day for 10 consecutive days to control mortality associated with columnaris disease caused by *F. columnare* in steelhead trout.
- Animals:** Approximately 76,800 steelhead trout juveniles.

- c. **Test article/controls:** The test article was a Type C medicated feed containing oxytetracycline that delivered a dose of 3.75 g oxytetracycline/100 pounds of fish/day for 14 consecutive days. The control was a non-medicated commercial salmonid feed.
- d. **Study Design:** Based on the clinical signs observed, columnaris disease was determined as the cause of increased mortality in seven tanks serving as the source of test fish. Test fish were systematically transferred to six test tanks. Untreated control and oxytetracycline-medicated groups were tested in triplicate. Treatments began the day following fish allocation to the study tanks. Fish were treated with medicated feed (3.75 g oxytetracycline/100 lbs of fish/day) for 14 consecutive days. Fish were observed and mortality recorded for an additional 10 days. Oxytetracycline administration was initiated in the control group twelve days after the start of the study due to the mortality rates in these tanks. The concentration of oxytetracycline in the medicated feed was assayed to verify the dosage.
- e. **Parameters Measured:** Mortality and water quality parameters were recorded.

Results: Mortality rates are included in the following table.

Table 2. Mortality rates for the 14-day treatment period and 10-day post-treatment period for a field study involving a columnaris infection in steelhead trout.

<u>Treatment Group</u>	<u>Mean Cumulative Mortality</u>	<u>Mean Percent Cumulative Mortality</u>
Control	1501	23.5
Oxytetracycline-treated	493	7.8

Conclusions: The results support the effectiveness of oxytetracycline-medicated feed administered at 3.75 g oxytetracycline/100 pounds of fish/day for 14 consecutive days for the control of mortality associated with columnaris disease, caused by *F. columnare*, in steelhead trout reared in freshwater.

3) Study Number BOZ-98-EFF-04

Title: “The Efficacy of Oxytetracycline-mediated Feed to Control Mortality of Juvenile Steelhead Trout *Oncorhynchus mykiss* Caused by Columnaris, Causative Agent *Flavobacterium columnare*, Study #04”

Study Director: James D. Bowker

Study Location: US Fish and Wildlife Service
Coleman National Fish Hatchery
Anderson, CA

General Design of the Study:

- a. Purpose: To evaluate the effectiveness of oxytetracycline-medicated feed administered at a dosage of 3.75 g oxytetracycline/100 pounds of fish/day for 10 consecutive days to control mortality associated with columnaris caused by *F. columnare* in steelhead trout.
- b. Animals: Approximately 32,120 steelhead trout juveniles
- c. Test article/controls: The test article was a Type C medicated feed containing oxytetracycline that delivered a dose of 3.75 g oxytetracycline/100 pounds of fish/day for 10 consecutive days. The control was a non-medicated commercial salmonid feed.
- d. Study Design: Fish from two of the four tanks serving as the source of test fish were examined. The body surface, fins, gills, and internal organs were examined and gill, kidney, spleen, and external skin lesion inocula were cultured and evaluated for the presence of systemic bacteria. Fish were originally held in four tanks. Fish from one tank were equally divided between two tanks. Fish were held in a total of eight tanks during the study. Four replicates were included in each treatment group. Treatment groups were untreated controls and oxytetracycline medicated. Treatments began the day following fish allocation to the study tanks. Fish were treated with medicated feed (3.75 g oxytetracycline/100 pounds of fish/day) for 10 consecutive days. Fish were observed and mortality recorded for an additional 14 days. The concentration of oxytetracycline in the medicated feed was assayed to verify the dosage.
- e. Parameters Measured: Mortality and water quality parameters were recorded.

Results: Mortality rates are included in the following table.

Table 3. Mortality rates for the 10-day treatment period and 14-day post-treatment period for a field study involving a columnaris infection in steelhead trout.

<u>Treatment Group</u>	<u>Cumulative Mortality</u>	<u>Mean Percent Cumulative Mortality</u>
Control	12,693	74.8
Oxytetracycline-treated	2,789	17.3

Conclusions: Results from this study support the effectiveness of oxytetracycline-medicated feed administered at a dosage of 3.75 g oxytetracycline/100 pounds of fish/day for 10 consecutive days to control mortality associated with columnaris caused by *F. columnare* in steelhead trout reared in freshwater.

4) Study Number BOZ-98-OTF-03

Title: “Efficacy of oxytetracycline-medicated Feed to Control Mortality of Fingerling Coho Salmon (*Oncorhynchus kisutch*) Caused by Coldwater Disease.”

Study Director: James D. Bowker

Study Location: US Fish and Wildlife Service
Quilcene National Fish Hatchery Road
Quilcene, WA

General Design of the Study:

- a. Purpose: To evaluate the effectiveness of oxytetracycline administered in feed at a dosage of 3.75 g oxytetracycline/100 pounds of fish/day for 10 consecutive days to control mortality associated with coldwater disease caused by *Flavobacterium psychrophilum* in coho salmon.
- b. Animals: Approximately 83,500 coho salmon fingerlings
- c. Test article/controls: The test article was a Type C medicated feed containing oxytetracycline that delivered a dose of 3.75 g/100 pounds of fish/day for 10 consecutive days. The control was a non-medicated commercial salmonid feed.
- d. Study Design: The test fish originated from a single concrete raceway. Ten fish from the raceway were examined (body surface, fins, gills, and internal organs), kidney inocula were cultured and examined for the presence of systemic bacteria, and stained kidney imprints examined microscopically for presence of systemic bacteria. Coldwater disease was determined to be the cause of increased mortality in the test fish. Test fish were randomly transferred to 12 test tanks. Untreated control and oxytetracycline-medicated groups were tested in replicates of six. Treatments began the day following fish allocation to the test tanks. Fish were treated with medicated feed (3.75 g oxytetracycline/100 pounds of fish/day) for 10 consecutive days. Post-treatment mortality was observed for an additional 14 days. The concentration of oxytetracycline in the medicated feed was assayed to verify the dosage.
- e. Parameters Measured: Mortality and water quality parameters were recorded.

Statistical Analysis: Mortality rates were analyzed using a t-test and the results confirmed using the GENMOD procedure in SAS[®].

Results: Mortality rates are included in the following table.

Table 4. Mortality rates for the 10-day treatment period and 10-day post-treatment period for a field study involving a coldwater infection in coho salmon.

Treatment Group	Mean Cumulative Mortality	Mean Percent Cumulative Mortality
Control	140.2	2.0
Oxytetracycline-treated	54	0.8
p-value		<0.0001

Conclusions: The results from this study demonstrate the effectiveness of oxytetracycline-medicated feed administered at a dosage of 3.7 g oxytetracycline/100 pounds of fish/day for 10 consecutive days to control mortality associated with coldwater disease caused by *F. psychrophilum* in coho salmon.

5) Coldwater Disease in All Salmonids

The effectiveness of oxytetracycline-medicated feed to control mortality in freshwater-reared salmonids due to coldwater disease, caused by *F. psychrophilum*, was demonstrated in a series of field trials conducted under INAD 009332. During six of sixteen studies with coldwater disease diagnosed by pathology, reductions in mortality were obtained at rates similar to those seen during Study BOZ-98-OTF-03 following the administration of oxytetracycline-medicated feed. During the ten other studies, reductions in mortality were observed, but the reductions were not as large as the reduction during Study BOZ-98-OTF-03. Oxytetracycline was administered at doses of 3.35 - 3.92 g/100 pounds of fish/day for 10 consecutive days during the six studies. Rainbow trout and cutthroat trout were the species of salmonids treated during the studies. The results of these studies demonstrate the effectiveness of oxytetracycline-mediated feed administered at a dose of 3.75 g oxytetracycline/100 pounds of fish/day for 10 consecutive days to control mortality in freshwater-reared salmonids due to coldwater disease caused by *F. psychrophilum*.

3. TARGET ANIMAL SAFETY:

No additional data were required. However, under INAD 009332 a series of twenty-nine field effectiveness trials were conducted during 1998, 1999 and 2000. During these trials cutthroat trout, rainbow trout and chinook salmon were treated for coldwater disease with oxytetracycline-medicated feed. Oxytetracycline was administered at doses of 3.35 – 3.96 g/100 pounds of fish/day for 10 consecutive days. Mortality data was collected prior to treatment, during treatment and after treatment. During twenty-seven of the twenty-nine trials mortality decreased during the 10-day treatment period and post-treatment period. No evidence of toxicity or adverse events was reported during the trials. The results of these studies provide additional data demonstrating that oxytetracycline-medicated feed administered at doses up to 3.75 g oxytetracycline/100 pounds of fish/day for 10 consecutive days is safe for freshwater-reared salmonids.