Date of Approval: December 24, 2003

FREEDOM OF INFORMATION SUMMARY

SUPPLEMENTAL NEW ANIMAL DRUG APPLICATION

NADA 130-435

OxyMarine™

oxytetracycline hydrochloride

To add a claim for the skeletal marking of finfish fry and fingerlings

Sponsored by:
Alpharma Inc.

1. *GENERAL INFORMATION*

a. File Number:	NADA 130-435
b. Sponsor:	Alpharma Inc. One Executive Drive P.O. Box 1399 Fort Lee, NJ 07024
c. Drug Labeler Code:	046573
d. Established Name:	oxytetracycline hydrochloride
e. Proprietary Name:	OxyMarine
f. How Supplied:	3.45 pound bags
g. How Dispensed:	over-the-counter
h. Amount of Active Ingredient:	1 gram per 2.73 grams of powder
i. Route of Administration:	immersion
j. Species/Class:	Finfish fry and fingerlings
k. Recommended Dosage:	200 to 700 mg oxytetracycline hydrochloride (buffered)/liter of water for 2 to 6 hours
l. Pharmacological Category:	Antimicrobial
m. Indications:	To mark skeletal tissues, most often the otoliths, of all finfish fry and fingerlings for subsequent identification.
n. Effect of Supplement:	To add a claim for the marking of skeletal marking of tissues of all finfish fry and fingerlings

2. *EFFECTIVENESS:*

A combination of data from many different fish species reared in different temperatures and management systems were used to support the determination of effectiveness in all teleost (bony) fish, consistent with the *Guidance for Industry: FDA Approval of Animal Drugs for Minor Uses and for Minor Species* (FDA/CVM January 1999). The range of oxytetracycline concentrations, 200 to 700 mg OTC/L of water, is supported by the studies summarized in this section, as well as the literature references at the end of this section.

a. Dosage Characterization:

Reports of successful marking of bony structures of fish, especially the otoliths, have been published for decades. There is a significant body of evidence that tetracyclines stain bony tissues in a wide range of species. The process of marking bony structures with tetracyclines was described in the literature as early as 1962. The literature reflects the widespread use of oxytetracycline by various routes and demonstrates the breadth and number of available publications on marking. This literature provided information to demonstrate the safety and effectiveness of oxytetracycline marking of finfish.

The otolith was selected for evaluation of marking success because otoliths are the first permanent calcified structures present in the earliest life stages of fish and are, effectively, biological internal tags. Once deposited, calcium in the otolith was mobilized little if at all.

Immersion was chosen as the route of administration because immersion marking allows fish to be mass marked with minimal handling. The doses selected were based on the doses found in the published literature.

b. Substantial Evidence:

1. Field Study

   Type of Study: Clinical Field Trial

   Name and Address of Investigator: W. Jenkins
   South Carolina Department of Natural Resources
   Charleston, SC

   General Design of the Study:

   a. Purpose of the Study: To evaluate the efficacy of using oxytetracycline (OTC) to mark red drum fingerlings for later identification as stocked fish in wild populations.

   b. Test Animals: Red drum fingerlings *(Sciaenops ocellatus)*

   c. Treatment Groups: This study included only one treatment group, oxytetracycline-treated fish.

d. Dosage Form: Water-soluble oxytetracycline hydrochloride

e. Route of Administration: Immersion (bath)

f. Dosages Used: 500 mg oxytetracycline/L of water for 4 hours

g. Test Duration: 30 days (treatment to examination)

h. Variables: Examination of otoliths to identify marks and mortality associated with OTC immersion marking

Methods: Red drum were harvested from 5 ponds in the fall of 1996 and 4 ponds in the spring of 1997 for immersion treatment. Fish were placed in a holding tank and acclimated to 15 ppt salinity water. OTC was added to the tank to provide a 500 mg/L OTC concentration. Fish were held for 4 hours. Fish were then transferred to a hauling trailer filled with 15 ppt salinity water. Fish were restocked in a pond at the culture facility and held for 7 days. The salinity in the pond was raised to the concentration of the area to be stocked during the 7-day holding period. Fish were harvested and transported to stocking sites for release. A subsample of each group of marked fish was retained for 30 days to confirm mark efficacy.

Results: Visible marks could be detected on otoliths in all batches of treated fish. Samples of fish from the wild population indicated that up to 40% of stocked age one year fish have OTC otolith marks.

Conclusion: Based on this study, the recommended dose of oxytetracycline to achieve a fluorescent mark on the otolith is a four-hour immersion at 500 mg/L oxytetracycline hydrochloride.

2. Field Study

Type of Study: Clinical Field Trial

Name and Address of Investigator: M. L. Hendricks
Pennsylvania Fish & Boat Commission
State College, PA

General Design of the Study:

a. Purpose of the Study: To apply unique multiple marks needed to discriminate between groups of fish from different egg source rivers, fish released as fry or fingerlings, and fish released at different sites.

b. Test Animals: American shad fry *(Alosa sapidissima)*, 3-15 days old

c. Treatment Groups: All fry produced received marks. Six different single and multiple marks were applied. A total of 8,500,000 fry were produced with all except 13,500 stocked in the Susquehanna River and its tributaries, or the Lehigh River. Fry were marked in 1200 L rearing tanks (40 total), each containing up to 500,000 fry.

d. Dosage Form: Water-soluble oxytetracycline hydrochloride

e. Route of Administration: Immersion (bath)

f. Dosages Used: 256 mg oxytetracycline/L of water for 4 hours

g. Variable: Fry from 6 tanks and raceways were sampled for otolith mark retention.

Results: Retention of immersion marks for American shad fry was 100% for all production groups in 1996. Refer to the following table.

Table 2.1. Oxytetracycline mark retention for American shad reared in 1996.

Tank/ Raceway	Mark Applied	Marks Visualized	Number Marked	Number Examined	Percent Marked	Number stocked
Race F1	9,12,15	9,12,15	19	19	100	171,700
Race F3	3,9,12,15	3,9,12,15	19	19	100	277,100
Race E1	3,6,9	3,6,9	18	18	100	42,900
Race F2	3,9,12	3,9,12	19	19	100	561,100
Race F4	3,6,9,12	3,6,9,12	19	19	100	682,500
Not Sampled	3	3	-	-	-	5,730,200
Tank J4*	3	3	17	17	100	Not stocked

*Sampled at 28 days of age.

Conclusion: Based on this study, the recommended dose of oxytetracycline to achieve a fluorescent mark on the otolith is a four-hour immersion at 256 mg/L oxytetracycline hydrochloride.

3. Field Study

Type of Study: Clinical Field Trial

Name and Address of Investigator: K. D. Cottrell
Illinois Department of Conservation
Springfield, Illinois

General Design of the Investigation:

a. Purpose of the Study: To determine the effectiveness of oxytetracycline hydrochloride in the marking of otoliths of fry (larval) and fingerling fish of different species.

b. Test Animals and Treatment Groups: Largemouth bass (*Micropterus salmoides*) fingerlings, walleye (*Stizostedion vitreum*) fry and fingerlings, and sauger (*Stizostedion canadense*) fry and fingerlings.

c. Treatment Groups: The fish were assigned to either treatment (500 mg/L for 6 hours) or smaller control groups.

Table 2.2. Number of each fish species treated during 1997.

Species	Treated	Control
Largemouth bass fingerlings	25,000	300
Walleye fry	5,837,400	100,000
Walleye fingerlings	143,306	1,000
Sauger fry	2,400,000	100,000
Sauger fingerlings	166,934	1,000

d. Dosage Form: Water-soluble oxytetracycline hydrochloride

e. Route of Administration: Immersion (bath)

f. Dosages Used: 0 and 500 mg oxytetracycline/L of water for 6 hours.

g. Test Duration: 30 to 45 days (from treatment to sampling).

h. Variables: Clinical observations and examination of otoliths to identify marks (50 fish from each treated group, and 50 from each control group).

Methods:

Fry – Groups of 100,000 fry were placed in a 500 mg/L concentration of oxytetracycline in 3 gallons of water within a plastic fish hauling bag for six hours. To ascertain mark retention, 100,000 of the marked and control fry were placed into separate rearing ponds. At the end of the rearing period (45 days for walleye and 60 days for sauger), samples of 50 fish per group were collected at harvest and checked for mark efficacy.

Fingerlings – Fish were immersed in a 500 mg/L concentration of oxytetracycline for six hours. Marked and control fish were retained for 30 days post-treatment and then harvested for examination. Samples of at least 50 fish from each group were checked for mark efficacy.

Results: Results are shown in the following table.

Table 2.3. Results of otolith marking field efficacy trials in 1997

Species	Treatment	Number Examined	Number Marked	Percent Marked
Walleye fry	OTC Immersion	50	50	100
	Control	50	7	14
Walleye fingerling	OTC Immersion	50	100	100
	Control	50	0	0
Sauger fingerling	OTC Immersion	50	50	100
	Control	50	0	0
Sauger fry	OTC Immersion	50	50	100
	Control	50	0	0
Largemouth bass fingerling	OTC Immersion	50	50	100
	Control	50	0	0

Conclusion: Based on this study, the recommended dose of oxytetracycline to achieve a fluorescent mark on the otoliths of walleye fry and fingerlings, sauger fry and fingerlings, and largemouth bass fingerlings is a six-hour immersion in 500 mg/L oxytetracycline hydrochloride.

4. Field Study

Type of Study: Clinical Field Trial

Name and Address of Investigator: D. O. Lucchesi
South Dakota Department of Game, Fish, and Parks
Pierre, South Dakota

General Design of the Study:

a. Purpose of the Study: To determine the effectiveness of oxytetracycline hydrochloride in the marking of otoliths of walleye fry (larval) and fingerlings.

b. Test Animals: Walleye (*Stizostedion vitreum*) fry and fingerlings.

Table 2.4. Fish treated during a field study.

Species	Number of Fish
Walleye fry	12,000,000
Walleye fingerlings	150,000

c. Treatment Groups: Natural rearing ponds were stocked with either 500 mg/L or 700 mg/L marked fry, or 500 mg/L marked fingerlings. Equal numbers of marked and unmarked fry were stocked into 0.8 ha hatchery ponds. Two ponds were stocked with 500 mg/L marked fry and

two ponds were stocked with 700 mg/L marked fry.

d. Dosage Form: Water-soluble oxytetracycline hydrochloride.

e. Route of Administration: Immersion (bath).

f. Dosages Used: Walleye fry were marked at either 500 mg/L or 700 mg/L. The fingerlings were marked at 500 mg/L. Immersion was for 6 hours.

g. Test Duration: 30 days to 3 months (from treatment to sampling).

h. Variables: Examination of otoliths to identify marks and mortality associated with OTC immersion marking

Methods:

To produce the marking bath, an OTC slurry was mixed in a 20 L plastic bucket and was then buffered to a neutral pH using sodium phosphate (dibasic, Na_2HPO_4). Walleye fry were immersed for 6 hours in 683 liter fiberglass raceways containing either 500 or 700 mg OTC/L. Density of fry did not exceed 2,000 fry per liter. Pond-reared fingerlings were marked by immersion for 6 hours in a 500 mg OTC/L water solution in 632 liter fiberglass transfer tanks. Fingerling densities did not exceed 50 fish per liter.

To evaluate marking efficacy and mark retention, OTC-marked fry and fingerlings were stocked into natural rearing ponds that had complete fish kills the previous winter. These test ponds contained marked individuals from one of three test groups: fry immersed in 500 mg OTC/L or 700 mg OTC/L, or fingerlings immersed in 500 mg/L. Test ponds were electrofished in early fall (approximately 3 months after marking) to recover walleye for evaluation of the mark quality.

To evaluate mortality associated with OTC immersion marking, the 0.8 ha ponds were seined approximately 30-40 days after stocking. A sample of 100 fingerlings per pond was examined for OTC otolith marks.

Results: Results are shown in the following table.

Table 2.5. Visibility of walleye otolith marks 3 months after marking as fry.

OTC Concentration (mg/L)	Mark Intensity			
	0	**1**	**2**	**3**
500[a]	50%	40%	10%	0
700[a]	0	0	18%	82%
700[b]	0	0	2%	98%

[a]1996 [b]1997

These findings suggest that better quality marks are obtained when fish are immersed in a bath containing 700 mg/L OTC. Problems with OTC precipitating out of solution typically occurred at a pH of higher than 8.0.

Therefore, the pH of the bath was maintained at 7.0 to 7.6, even when hatchery water pH exceeded that range.

Conclusion: Based on this study, the recommended dose of oxytetracycline to achieve a fluorescent mark on the otolith of walleye fry is a six-hour immersion at 700 mg/L OTC.

SUPPORTING LITERATURE

The following published articles were submitted to support the effectiveness of oxytetracycline for the marking of otoliths in bony fish:

1. Brooks, R.C., R.C. Heidinger, and C.C. Kohler. Mass-Marking Otoliths of Larval and Juvenile Walleyes by Immersion in Oxytetracycline, Calcein, or Calcein Blue. North American Journal of Fisheries Management 14: 43-150, 1994.

2. Hendricks, M.L., Bender, T.R., and Mudrak, V.A. Multiple Marking of American Shad Otoliths with Tetracycline Antibiotics. North American Journal of Fisheries Management, 11:212-219, 1991.

3. Secor, D.H., M. G White, and J.M Dean. Immersion Marking of Larval and Juvenile Hatchery-Produced Striped Bass with Oxytetracycline. Transactions of the American Fisheries Society, 120:261-266, 1991.

4. Thomas, L.M. Chemical Mark Application in Red Drum (*Sciaenops ocellatus*). Thesis for Master of Science at Corpus Christi State University, Corpus Christi, Texas. 44 pages, 1993.

5. Younk, J.A., Cook, M.F. Fluorescent Chemical Marking of Walleye Larvae with a Selected Literature Review of Similar Investigations. Minnesota Department of Natural Resources Investigational Report 408. 18 pages, 1991.

3. TARGET ANIMAL SAFETY:

A. "Toxicity of Oxytetracycline and Calcein to Juvenile Striped Bass."

1. Type of Study: Target animal safety

2. Name and Address of Investigators: B. W. Bumguardner and T. L. King
Texas Parks and Wildlife Department
Palacios, Texas

3. General Design of the Study:

a. Purpose of the Study: The study was designed to identify adverse effects of exposing fish to a geometric sequence of oxytetracycline by immersion.

b. Test Animals: Juvenile striped bass (*Morone saxatilis*), approximately 48 mm total length and 2.2 g.

c. Treatment Groups: 3 replicates per concentration tested. 10 fish per replicate. The first set of tests included 6 different OTC concentrations. The second set of tests included 3 higher concentrations.

d. Dosage Form: Water-soluble oxytetracycline hydrochloride

e. Route of Administration: Immersion (bath).

f. Dosages Used: Concentrations of 0, 55.8, 111.6, 223.3, 446.5, and 893 m/L; second set of tests: 893, 1786, and 3572 mg/L. 6 hour exposure.

g. Test Duration: 26 days (from set up to final observations).

h. Variables: Survival and behavior at 1 hour intervals during the 6 hour treatment and for 6 hours after treatment. Mortality was monitored daily for the next 4 days. Water conditions were also monitored for temperature, salinity, pH, and total hardness.

4. Methods:

After a 22-day acclimation period in a recirculating raceway, the fish were transferred to the test chambers. The test chambers were 4.5 liter aquaria containing salt water (enough to make the total volume after addition of OTC solution 2 liters). OTC HCl unbuffered solution was added to provide concentrations of 0, 55.8, 111.6, 223.3, 446.5, and 893 mg/L. Since no deaths were seen, the 893 mg/L concentration was repeated and 1786 and 3572 mg/L concentrations were added. Fish were immersed for 6 hours. Fish were then placed in a beaker while the aquaria were rinsed. Fish were then placed back into aquaria. Water was exchanged every 24 hrs for 4 days.

5. Results:

a. Water conditions: Water conditions are included in the following table.

Table 3.1. Water conditions during a study to evaluate the safety of calcein and oxytetracycline solutions with juvenile striped bass.

Condition	Pre Treatment	During Treatment
Temperature	26°C	26°C
Salinity	5 ± 1.9%	7%
pH	8.3	3.25-8.56*
Total hardness		33.4 mg/L (as CaCO3)

*pH varied depending on OTC concentration

A white precipitate was observed in the higher OTC concentration test chambers.

b. Mean Mortality: Percent mean mortality results are included in the following table.

Table 3.2. Mean mortality (%) results of a study to evaluate the safety of calcein and oxytetracycline solutions with juvenile striped bass.

Concentration	During Treatment	After Treatment	Total (and range)
0 (control)	0	6.7	6.7 (0-10)
55.8 mg/L	0	0	0
111.6 mg/L	0	0	0
223.3 mg/L	0	0	0
446.5 mg/L	0	30	30 (10-60)
893 mg/L	0	81.7	81.7 (60-100)
1786 mg/L	23.3	73.3	96.7 (90-100)
3572 mg/L	100	-	100

Stressed behavior (rapid swimming at the water's surface) was reported in concentrations of OTC 446.5 mg/L and higher.

6. Conclusion: The pH of the immersion solution may have caused or contributed to the death of the fish in the highest dose groups. The mean pH of test aquaria water after addition of OTC solution began to decrease significantly at 893 mg OTC/L. At 1786 mg/L, the pH was more than 1.5 units lower than that of the control group; at 3572 mg/L, the pH was more than 5 units lower than that of the control group

   Water quality, especially pH, must be monitored and controlled when using higher concentrations of OTC for immersion of fish. Buffering of the water can be done to maintain a healthy pH when OTC is used.

B. Field Study

1. Type of Study: Clinical Field Trial

2. Name and Address of Investigator: W. Jenkins
   South Carolina Department of Natural Resources
   Charleston, SC

3. General Design of the Study:

   a. Purpose of the Study: This study was primarily designed to demonstrate effectiveness. During the fall of 1996 and spring of 1997, fish were stocked in hatchery ponds following treatment. The ponds were harvested approximately one week later to evaluate post-treatment mortality.

   b. Test Animals: Red drum *(Scianenops ocellatus)* fingerlings.

   c. Treatment Group: This study included only one treatment group, oxytetracycline-treated fish.

   d. Dosage Form: Water-soluble oxytetracycline hydrochloride

   e. Route of Administration: Immersion (bath)

   f. Dosages Used: 500 mg oxytetracycline/L for 4 hours

   g. Test Duration: Approximately one week (treatment to harvest)

   h. Variables: Mortality occurring one week following treatment was recorded.

4. Methods: Red drum fingerlings were placed in a holding tank on a trailer and acclimated to approximately 15 ppt salinity. Adequate OTC was added to provide a 500 mg/L concentration. Fish were held for 4 hours and then transferred to a hauling trailer. Fish were stocked in a pond at the culture facility and held for one week. During that week pond salinity was slowly raised to the same concentration as the area to be stocked (29-30 ppt). Fish were then harvested and transported to stocking sites for release.

5. Results: Results are included in the following table.

Table 3.3. Mean size and survival of red drum fingerlings treated from 9/96-5/97 with OTC at a concentration of 500 mg/L for 4 hours.

Treatment Date	Mean Length (mm)	Mean Weight (g)	Number of Fish Treated	Survival (%)	Number Harvested
9/16	39	0.59	37,047	72.2	26,746
10/15	37	0.67	63,898	43.1	28,061[a]
10/16	25	0.21	381,028	66.6	253,637
10/22	37	0.68	43,332	71.1	30,801
11/7	13	0.10	265,593	86.4	229,712[b]
5/15	33	0.36	65,810	82.8	54,474
5/22	28	0.25	114,423	80.7	92,395
5/27	29	0.24	470,611	86.8	408,653
5/29	34	0.31	143,589	93.9	134,900

[a]This pond had a dense growth of macrophytic algae. Fish got stranded in the algal mat during harvest.

[b]A salinity acclimation error during post-treatment resulted in 20% mortality during harvest and release, therefore only 183,770 fish were released alive.

6. Conclusion: Based on this study, the recommended dose of oxytetracycline to achieve a fluorescent mark on the otolith, a four-hour immersion in 500 mg/L oxytetracycline, is safe to red drum fingerlings.

C. Field Study

1. Type of Study: Clinical Field Trial

2. Name and Address of Investigator: K. D. Cottrell
Illinois Department of Conservation
Springfield, Illinois

3. General Design of the Study:

a. Purpose of the Study: The studies were primarily designed to demonstrate effectiveness. The studies involved the treatment of many species and large numbers of fish. The studies demonstrate several issues related to target animal safety.

b. Test Animals: Largemouth bass (*Micropterus salmoides*) fingerlings, walleye (*Stizostedion vitreum*) fry and fingerlings, and sauger (*Stizostedion canadense*) fry and fingerlings.

c. Treatment Groups: The fish were assigned to either treatment (500 mg/L for 6 hours) or smaller control groups according to the following table.

Table 3.4. Treatment groups for a marking study conducted in 1997.

Species	Treatment (number)	Control (Number)
Largemouth bass fingerlings	25,000	300
Walleye fry	5,837,400	100,000
Walleye fingerlings	143,306	1,000
Sauger fry	2,400,000	100,000
Sauger fingerlings	166,934	1,000

d. Dosage Form: Water-soluble oxytetracycline hydrochloride

e. Route of Administration: Immersion (bath).

f. Dosages Used: 0 and 500 mg oxytetracycline/L of water for 6 hours

g. Test Duration: 1 day (fish stocked the day of treatment)

h. Variables: Mortality and adverse reactions were recorded.

4. Methods:

Fry - Groups of 100,000 fry were placed in a 500 mg/L concentration of oxytetracycline in 3 gallons of water within a plastic fish hauling bag for six hours.

Fingerlings - Fish were immersed in a 500 mg/L concentration of oxytetracycline for six hours.

5. Results: Results are included in the following table.

Table 3.5. Mortality and adverse reactions observed in 1997 at Jake Wolf, Little Grassy and LaSalle Hatcheries in Illinois.

Species	Number Treated	Mortalities	Adverse Reactions
Largemouth bass fingerlings	25,000	0	None
Walleye fry	5,824,080	0	None
Walleye fingerlings	138,626	0	None
Sauger fry	22,000,000	0	None
Sauger fingerlings	166,934	0	None

No adverse reactions or mortality occurred in control groups in any of the species treated.

6. Conclusion: Based on this study, the recommended dose of oxytetracycline to achieve a fluorescent mark on the otolith, a six-hour immersion at 500 mg/L, is safe to fry and fingerlings

D. Field Study

1. Type of Study: Clinical Field Trial

2. Name and Address of Investigator: R. T. Colesante
New York State Department of
Environmental Conservation
Constantia, NY

3. General Design of the Study:

a. Purpose of the Study: Evaluate the survival of walleye fry after oxytetracycline (OTC) marking and stocking in earthen ponds.

b. Test Animals: Walleye (*Stizostedion vitreum*) fry

c. Treatment Groups: Oxytetracycline-treated and untreated fry.

d. Dosage Form: Water-soluble oxytetracycline hydrochloride

e. Route of Administration: Immersion (bath)

f. Dosages Used: 0 and 500 mg oxytetracycline/L of water for 6 hours

g. Test Duration: 45-55 days (treatment to harvest)

h. Variable: Survival was recorded.

4. Methods: In 1994, fry were immersed in a 500 mg/L oxytetracycline static bath for 6 hours or left untreated. Six ponds were stocked with marked fry and an additional six ponds were stocked with unmarked fry. Each pond was stocked with 20,000 fry. Ponds were harvested after 45-55 days.

5. Results: Survival results from each pond are shown in the following table.

Table 3.6. Survival results following otolith marking with oxytetracycline.

Pond No.	Treatment Group	Fry Stocked	Fingerling Return	
			Unmarked	Marked
1	OTC	20,000	-	22,728
2	OTC	20,000	-	15,557
3	OTC	20,000	-	11,347
4	Control	20,000	5,360	-
5	Control	20,000	12,964	-
6	OTC	20,000	-	14,118
7	Control	20,000	18,184	-
8	OTC	20,000	-	13,432
9	Control	20,000	12,367	-
10	Control	20,000	12,727	-
11	Control	20,000	13,457	-
12	OTC	20,000	-	11,697
Mean Survival			**62.5 %**	**74.1 %**

6. Conclusion: Based on this study, the recommended dose of oxytetracycline to achieve a fluorescent mark on the otolith, a six-hour immersion at 500 mg/L, is safe to walleye fry.

4. HUMAN SAFETY:

- Toxicity - An acceptable daily intake (ADI) of 25 micrograms per kilogram of body weight per day has been previously codified for total tetracycline residues (chlortetracycline, oxytetracycline, and tetracycline) (21 CFR 556.500).

- Residue Depletion Studies - Residue depletion data for fish marked with oxytetracycline have been summarized in PMF 5667 (67 FR 46527). The data are from PMF 3265 and the public literature. A discussion on the long inherent withdrawal period which occurs between treatment of the fish and possible consumption by humans also appears in PMF 5667. The data in PMF 5667 and the public literature, and the long inherent withdrawal period support the human food safety of the use of oxytetracycline to mark finfish fry and fingerlings.

- Tolerance and Withdrawal Time - A tolerance of 2 ppm in muscle tissue as the sum of tetracycline residues has been previously codified for the edible tissue of salmonids and catfish (21 CFR 556.500). This tolerance is now being extended to all finfish. A withdrawal time beyond the grow-out period is not needed.

- Microbial Food Safety - The potential human health impact of the microbial effects associated with the use of oxytetracycline HCl to mark skeletal tissue, most often the otoliths, of finfish fry or fingerlings for subsequent identification, was assessed pursuant to CVM's Guidance for Industry #78 titled *Consideration of the Human Health Impact of the Microbial Effects of Antimicrobial New Animal Drugs Intended for Use in Food-Producing Animals*. The Agency has determined that use of oxytetracycline HCl as described in this application will not significantly impact the rate and extent of development of antimicrobial drug resistant enteric bacteria formed in the intestinal tract of treated fish following exposure to oxytetracycline HCl.

- Regulatory Method for Residues - The analytical method for detection of residues of oxytetracycline is a microbiological assay using *Bacillus cereus* var. *mycoides*. This method may be found in "Antibiotic Residues in Milk, Dairy Products, and Animal Tissues: Methods, Reports, and Protocols" (revised October 1968, reprinted December 1974), National Center for Antibiotic and Insulin Analysis, FDA, Washington, DC 20204). The method is on file at the Center for Veterinary Medicine, 7500 Standish Pl., Rockville, MD 20855.

5. AGENCY CONCLUSIONS:

The data submitted in support of this NADA satisfy the requirements of section 512 of the Federal Food, Drug, and Cosmetic Act and 21 CFR Part 514 of the implementing regulations. The data demonstrate that OXYMARINE, when administered by immersion at concentrations of 200 to 700 mg oxytetracycline hydrochloride/liter of water for 2 to 6 hours, is safe and effective to mark skeletal tissues, most often the otoliths, of all finfish fry or fingerlings for subsequent identification.

Oxytetracycline Soluble Powder for use in food-producing animals is currently marketed as an over-the-counter product. Adequate directions for safe and effective use by the layperson have been provided. Therefore, the Center for Veterinary Medicine has concluded that this product retains over-the-counter status.

A tolerance of 2 ppm in muscle tissue as the sum of tetracycline residues has been previously codified for the edible tissue of salmonids and catfish (21 CFR 556.500). This tolerance is now being extended to all finfish. A withdrawal time beyond the grow-out period is not needed. An acceptable daily intake (ADI) of 25 micrograms per kilogram of body weight per day has been previously codified for total tetracycline residues (chlortetracycline, oxytetracycline, and tetracycline). The potential human health impact of the microbial effects associated with the use of oxytetracycline HCl to mark skeletal tissue as described in this document was assessed. The Agency has determined that use of oxytetracycline HCl as described in this application will not significantly impact the rate and extent of development of antimicrobial drug resistant enteric bacteria formed in the intestinal tract of treated fish following exposure to oxytetracycline HCl.

Under section 512(c)(2)(F)(iii) of the Federal Food, Drug, and Cosmetic Act, this approval for food-producing animals does not qualify for marketing exclusivity.

In accordance with 21 CFR 514.106(b)(2)(vii), this is a Category II change involving the addition of a species and a new claim with a separate dose and treatment regimen. The safety and effectiveness data in the parent application did not need to be reevaluated.