FILE

FINDING OF NO SIGNIFICANT IMPACT

for

Romet^{R}-30 Medicated Premix for Salmonid Fish (Trout and Salmon)

NADA 125-933
Hoffman-LaRoche Inc.

The Center for Veterinary Medicine has carefully considered the potential environmental impact of this action and has concluded that this action will not have a significant effect on the quality of the human environment and that an environmental impact statement therefore will not be prepared.

Hoffman-LaRoche Inc. of Nutley, New Jersey, has filed a new animal drug application (NADA 125-933) providing for the use of Romet-30 medicated premix at a variable concentration of about 0.1 to 0.5% in the feed for the control of furunculosis (caused by Aeromonas salmonicida), an infectious bacterial disease of salmonids. Fish are to be dosed at a level of 50 mg of drug active ingredient(a.i.)/Kg fish body weight. The feed medicated with Romet-30 for salmonids is to be used continuously for a maximum of five days. Salmonids given Romet-30 under these conditions cannot be used as food for a period of six weeks after termination of treatment.

Romet-30 is a feed premix which has a broad spectrum antibacterial and anticoccidial activity. This activity is due to the two active drug ingredients, sulfadimethoxine and ormetoprim, present as 25% and 5%, respectively, of the Romet-30 premix. Sulfadimethoxine is a sulfonamide antibiotic which has been widely used in the treatment of a variety of infectious diseases in humans and in domestic animals. Ormetoprim is a pyrimidine which is used primarily to potentiate the antibacterial and anticoccidial activity of sulfadimethoxine.

Rofenaid^{R}-40 premix is another combination of these active drug ingredients (a premix containing 25% sulfadimethoxine and 15% ormetoprim) which has already been approved for use in the prevention and treatment of several infectious diseases in chickens, turkeys and ducks.

Hoffman-LaRoche has filed the attached environmental impact analysis report (EIAR, pp. 1-98, dated February 22, 1984) in support of the proposed use of Romet-30 in salmonids. Portions of this EIAR are taken verbatim from the EIAR (dated January 31, 1983) submitted for the use of Rofenaid-40 in ducks (NADA 40-209).

The EIAR (attached) asserts that the proposed uses of Romet-30 in salmonids should result in minor increases in the introductions of sulfadimethoxine and ormetoprim into the environment, and that the levels of these two active ingredients expected in the environment should not result in effects on organisms in the environment. These assertions are based upon data and

be at least 95 million pounds of salmonids/year.

2. Environmental introduction of chemicals due to the manufacture of Romet-30:

The attached EIAR only partially estimates the increase in environmental introductions of chemicals that could result from the increased manufacture of both sulfadimethoxine and ormetoprim. Using even the optimistic "worst-case" assumptions made in the EIAR (20 million pounds of fish, treated once for 5 days) would result in the manufacture of an additional 2,273 Kg of these two active ingredients (1,894 Kg of sulfadimethoxine and 379 Kg of ormetoprim). The EIAR dated January 21, 1983 (Rofenaid-40 for ducks, NADA 40-209) estimated that 72,730 Kg of Rofenaid-40 premix (29,000 Kg of active drug ingredients) was sold in 1982. Use of Romet-30 in salmonids could therefore represent an increase in annual production of these two drug active ingredients of about 10%/year.

Production material balance data were given on p. 52 of the attached EIAR. This data can be used for an estimate of the potential annual increase in the chemical pollutants emitted into the environment due to the production of each additional kilogram of either sulfadimethoxine or ormetoprim:

	Chemical Waste from Producing 1 Kg Sulfadimethoxine	Annual Increase in Sulfadimethoxine Production	Increased Annual Chemical Emission
Solids disposal	0.361 Kg	1,894 Kg	683 Kg
Air discharges	0.21 Kg	1,894 Kg	398 Kg
Water discharges	4.561 Kg	1,894 Kg	8,639 Kg

	Chemical Waste from Producing 1 Kg Ormetoprim	Annual Increase in Ormetoprim Production	Increased Annual Chemical Emission
Liquids disposal	6.468 Kg	379 Kg	2,450 Kg
Solids disposal	1.011 Kg	379 Kg	383 Kg

Air emissions for ormetoprim were not included in the material balance data, however, air emissions for ormetoprim do apparently occur (listed on p. 53 of the attached EIAR).

3. Environmental introductions of chemicals due to the of Romet-30:

Romet-30 is to be used for the prevention and therapy of furunculosis in salmonid fish. The main route of introduction of sulfadimethoxine and ormetoprim (and their metabolites) into the environment therefore appears to be through the excretion of food wastes into water by the medicated animals. The EIAR, on pp. 47-50, estimated that the maximum amount of total drug active ingredients that might be found in the water of the raceway where fish had been treated for five days should range from about 20 to 60 parts per billion (ppb). This means that the maximum raceway water levels of sulfadimethoxine could range from 17-51 ppb and ormetoprim from 3-9 ppm.

An experiment was performed to assess the accuracy of this use estimate (pp. 49-50 and appendices on pp. 60-97 of the attached EIAR). Rainbow trout were kept in a model raceway and the trout were given an appropriately medicated feed for five days. Water samples were taken several times a day during the five days of treatment and for the following three days. Samples of the sediment (wastes) in the raceway were also taken on the fourth day after treatment had stopped. Samples of water and sediments (which had been filtered) were subsequently analysed for levels of sulfadimethoxine and ormetoprim (detection limits of 0.5 ppb). Considerable variability was seen in the levels of sulfadimethoxine and ormetoprim found in the samples of water. The highest level of sulfadimethoxine found in the water was 25 ppb on the second day of fish treatment. The highest level of ormetoprim found in the water was 6 ppb and this was also found on the second day of drug therapy. These measured values support the above estimates of maximum raceway drug concentrations. Therefore these two measured values (of 25 and 6 ppb) appear to be reasonable worst-case estimates for levels of sulfadimethoxine and ormetoprim that could be expected in the water coming from the raceways of trout appropriately treated with Romet-30.

Examination of the raceway sediment (fish wastes and perhaps uneaten food) clearly indicates that much higher levels of these drugs are likely to be found in this component of the raceway environment. Levels of sulfadimethoxine in the sediment and its filtrate averaged 255 ppm and 9 ppm, respectively. Levels of ormetoprim in the sediment and its filtrate averaged 7.4 ppm and 0.7 ppm, respectively. Disposal of raceway sediment after drug treatments with Romet-30 is one of the potentially more serious environmental problems from the use of this drug.

B. Fate of Sulfadimethoxine and Ormetoprim in the Environment

The fate of a chemical in the environment is influenced by the physical-chemical properties of the introduced chemical as well as the physical-chemical and biological properties of the sites of introduction. In broad terms, the fate of the introduced chemical is a function of the biological and chemical transformations that may occur and the transport of the introduced chemical and/or degradation products away from the site of introduction.

Data are limited on the environmental fate of sulfadimethoxine and ormetoprim. Important physical-chemical data that are commonly used to predict the partitioning of chemicals among environmental components (such as vapor pressure, dissociation constant, octanol to water partitioning coefficient, and Freundlich soil sorption coefficient), were not found in this application or in previous approvals for either sulfadimethoxine or ormetoprim. Such information would help firmly establish how likely either of these two chemicals would be to partition into the atmosphere or from water (or sediment) into living things.

A chemicals low solubility in water is often strongly correlated with the chemical being tightly bound to soil (Kenaga and Goring, 1980). Low water solubility is also strongly correlated with a chemicals ability to bioaccumulate (partition into lipids). The attached EIAR mentions that the solubility of sulfadimethoxine in water is 50 mg/L and that of ormetoprim is 200 mg/L.

Ormetoprim should therefore more readily partition (than would sulfadimethoxine) from soil into the aquatic environment. However, the EIAR (on pp. 17-24) describes a soil study in which water was leached through three different soil types. This study appears to indicate that water leaching through soils removed about 50 to 80% of the sulfadimethoxine, but none of the ormetoprim.

In addition, based on its water solubility, ormetoprim should be less soluble in lipids and therefore have less ability to bioaccumulate than would sulfadimethoxine. However, the EIAR (on pp. 10-11) describes a tissue residue study result inconsistent with this expectation. Trout were given medicated feed so that fish received drug at 50 mg/Kg (b.w.)/day for five days. The drug tissue residues were subsequently monitored for 13 weeks. Sulfadimethoxine tissue residues increased while the fish were being dosed, but were not detectable (< 0.05 ppm) within a few days after treatment was stopped. Ormetoprim levels in tissues also increased during the drug dosing, markedly so in the lipid rich fish skin. The ormetoprim levels in several fish tissues depleted much more slowly than did sulfadimethoxine. Residues of ormetoprim were still detectable in fish skin and scales at the end of this 13-week experiment. On that basis, a six-week withdrawal period was determined to allow a sufficient depletion of drug residues from the edible tissues.

Comparison of the results of these two studies with the water solubility data given indicate that further clarification on some of the physical-chemical properties and the environmental fate of these two drugs is highly desirable. As mentioned above, definitive measurements of the vapor pressure, dissociation constant, octanol to water partitioning coefficient, and Freundlich soil sorption coefficient would help determine the potential fate of these two drugs in the environment.

The stability of sulfadimethoxine and ormetoprim in the aquatic environment is evaluated on pp. 5-7 and 16-20 of the attached EIAR. Sulfadimethoxine and ormetoprim often both appear to be very stable in aqueous solutions. Exposure of solutions of each drug to high intensity ultraviolet light for 24 hours resulted in no degradation of either compound. Therefore neither sulfadimethoxine, nor ormetoprim would appear to be subject to significant photodegradation in the environment by sunlight.

Stability studies were also done with duck wastes mixed 1 to 20 with water. Such mixtures were fortified with either sulfadimethoxine or ormetoprim and kept basically under aerobic or anaerobic conditions. The levels of active drug ingredients in the water were analyzed for up to 55 days. Under aerobic conditions (i.e., with aeration), the levels of both drugs decreased relatively rapidly with half-lifes of about 7 and 10 days for sulfadimethoxine and ormetoprim, respectively. Under anaerobic conditions, the levels of both drugs decreased to approximately 50% within about 7 and 14 days for sulfadimethoxine and ormetoprim, respectively. However, no further decrease in either drug concentration was seen over the rest of this 55-day test. Depending on the environmental conditions, it therefore appears that each of these drugs may or may not be stable in the aquatic environment. Therefore specific information on the mechanisms and processes of degradation (e.g., chemical, biological, etc.) of these drug molecules and whether degradation products retain antimicrobial activity is desirable.

The two "worst-case" raceway estimates made on pp. 47-49 of the EIAR assume that all of the drug active ingredients are available (i.e., stable). The model raceway study verified that the highest drug levels reached under actual use conditions (25 and 6 ppb for sulfadimethoxine and ormetoprim, respectively) were reasonably encompassed within the drug ranges estimated in the "worst-case" calculations (17-51 ppb for sulfadimethoxine and 3-9 ppb for ormetoprim). Therefore, even if these two drugs are stable, the concentrations reached in water appear to be low enough to probably be of little concern to organisms in the aquatic environment (see below). The concentrations of sulfadimethoxine and ormetoprim found in fish wastes, however, (255 and 7.4 ppm, respectively) may be of more concern, especially if these two compounds are stable in such wastes.

C. Effects of Sulfadimethoxine and Ormetoprim in the Environment

The use of Romet-30 in salmonids is going to essentially restrict the introduction of sulfadimethoxine and ormetoprim into the aquatic environment. Unless sulfadimethoxine and/or ormetoprim partition into the atmosphere, it is reasonable to assume that the potential environmental effects of these two compounds should be restricted to organisms actually living in the water columns, or to those organisms living in the sediments contaminated by this water or by any raceway wastes released into the aquatic environment. Comparisons are made below between the levels of these two drugs that might be expected to occur in the aquatic environment and the drug levels that might be expected to cause effects in the few aquatic organisms that have actually been tested for the toxic effects of these two drugs. The potential for effects on other organisms in the environment is also subsequently briefly discussed below.

1. Aquatic organisms in the water column:
The attached EIAR, on pp. 38-41, briefly describes the aquatic toxicity tests that have been performed using sulfadimethoxine, ormetoprim and the two drug combination premixes Rofenaid-40 and Romet-30. Sulfadimethoxine, ormetoprim and Rofenaid-40 were tested in water for effects (EC50 or LC50) on three freshwater organisms: a green alga (Selenastrum capricornutum), a crustacean (water flea, Daphnia magna), and a fish (bluegill, Lepomis macrochirus). The two active drug ingredients and Romet-30 were tested in the feed for effects (LC50) on two freshwater fish species: rainbow trout, (Salmo gairdneri) and channel catfish (Ictalurus punctatus). The results of these studies are briefly summarized in Table 1 below.

Table 1. The 96-hour LC50 (or EC50) values (95% Confidence Intervals) of sulfadimethoxine (S), ormetoprim (O), Rofenaid-40 (R-40) and Romet-30 (R-30) to five freshwater organisms.

	Drug Concentration (mg/L or mg/Kg = ppm)			
	S	O	R-40	R-30
Selenastrum EC50 (decreased chlorophyll)	170(42-688)[b]	90(21-378)	38(6-238)	
Daphnia EC50 (48 hours) (immobilization)	53(26-105)	33(18-60)	38(23-61)	
Lepomis LC50	--[c]	--[c]	--[c]	
Salmo LC50[a]	400[d]	400[d]		400[d]
Ictalurus LC50[a]	400[d]	200(163-245)		600(378-952)

a = Drug dose given in the feed (mg/Kg fish body weight)
b = Brackets contain 95% confidence interval
c = No mortalities seen at highest possible concentration in water (S = 50 ppm, O = 200 ppm)
d = Highest concentration in feed resulting in no mortalities

Complete study reports examining the acute toxic effects of these compounds in water on a green alga, the water flea and bluegill were submitted to the NADA for review and these studies generally appeared to have been performed adequately. However, reports for the studies measuring the acute toxic effects of these compounds given in the feed on rainbow trout and channel catfish have not been submitted to the NADA for review. Only limited summary information from these studies was present in NADA 125-933. Therefore the adequacy of the feeding toxicity studies cannot be evaluated.

The data in Table 1 are the results of tests of the short-term toxicity of these compounds to five aquatic organisms. An LC50 (or EC50) is an estimated concentration that would, on the average, cause mortality (or some equivalent effect) in 50% of a population of organisms. The 95% confidence interval is the range of values within which it is 95% certain that the true LC50 (or EC50) resides. Contrary to the information in the attached EIAR, the 95% confidence interval does not delineate either the lower or upper range of mortality (or equivalent effect).

A key value not reported in any of these studies is the slope of the dose-response line. From the slope of the line, scientists determine the rapidity of the onset of the adverse effect as dose increases. The steeper the slope of the line, the more suddenly mortalities occur with increasing increments of exposure. It is from this information that the maximum concentration where the effect would not be observed is estimated.

Depending on the effect being measured in the test and the slope of the dose-response line, a safety factor is applied to the study results and a safe concentration is estimated where the test organism (and the class of organisms it respresents) can be expected to grow, reproduce, and behave normally.

On average, the safety factor is one-one hundredth of the 50% effect concentration when the effect is mortality or its equivalent (LC50 or EC50). This safety factor is frequently used when adequate data are not available to determine a more accurate value.

In Section A.3. above, the measured maximum concentration of sulfadimethoxine and ormetoprim in water were found to be 25 ppb and 6 ppb, respectively. If a safety factor of 1/100 is applied to the reliable aquatic acute toxicity data in Table 1, a comparison can be made with these measured concentrations (Table 2).

Table 2. Comparison of maximum sulfadimethoxine (S), ormetoprim (O), and Rofenaid-40 (R-40) concentrations with estimated safe concentrations for aquatic organisms.

Measured maximum concentrations in raceway water	Estimated "safe" concentrations of S, O, and R-40 based on acute aquatic toxicity data in Table 1*			
		S	O	R-40
S = 25 ppb	Selenastrum	1,700 ppb	900 ppb	300 ppb
O = 6 ppb	Daphnia	530 ppb	330 ppb	380 ppb
S+O Combination = 31 ppb	Lepomis	--	--	--

*One-one hundredth safety factor applied to reliable aquatic acute toxicity data

From this comparison, we can generally conclude that the drug components of Romet-30 would appear unlikely to cause acute adverse effects on representative algae, fish or invertebrates that exist in water which comes directly from a raceway treated with this drug. Division of the "safe" concentrations by the measured drug concentrations illustrates that moderate margins of protection (of from about 12-150 X the safe concentration) exist and these margins of protection should thereby prevent effects of these drugs on these aquatic organisms.

No information relevant to predicting the effects of sulfadimethoxine or ormetoprim or Romet-30 on specific strains of bacteria, free-living protozoans, or fungi could be found. Where these two compounds have been claimed to have a broad spectrum of activity against pethogenic bacteria and protozoa (coccidia), it would be surprising if similar organisms in the aquatic environment were not sensitive to these active drug ingredients. A screening for the inhibitory concentrations of these two compounds (and their combination) on bacteria, free-living protozoans, and fungi would seem highly desirable.

2. Aquatic organisms in sediments:
There appears to be no information available for predicting the effects of any of the individual drug components (or a combination of them) on sediment (or soil) bacteria, protozoans, fungi, benthic crustaceans, worms, clams, snails or rooted aquatic macrophytic plants. Aquatic sediments appear to be one environment where drug effects information may be necessary. Especially as the highest concentrations of sulfadimethoxine and ormetoprim in the aquatic environment (255 ppm and 7.4 ppm, respectively) are likely to be found in the raceway wastes after the use of Romet-30 in trout. It appears likely that such levels of sulfadimethoxine and ormetoprim could have an effect on at least some of the bacteria and protozoans so abundant at the interface between the aquatic sediments and water.

Raceway sediments may be handled in a fashion to reduce these drug levels (e.g., degradation via sewage treatment). Even if such fish wastes are released directly into the environment, they could be scattered throughout a fairly large area of sediment and thereby be distributed fairly widely in the river or stream receiving these wastes. Any effects that might result would perhaps be expected to be of an intermittent and short-term nature. Such effects would be located in the immediate tributaries receiving such waste and dilution with water from adjacent streams could also commonly occur.

3. Terrestrial organisms:
It appears unlikely that significant quantities of either sulfadimethoxine or ormetoprim will be introduced into the terrestrial environment as a result of the use of Romet-30 in salmonids. The attached EIAR (on pp. 8-10, 13, and 29-32) briefly summarizes studies demonstrating that sulfadimethoxine, ormetoprim, and Rofenaid-40 are not very acutely toxic to six species of terrestrial plants, and are also not very acutely (or chronically) toxic to two avian species or four mammalian species. It appears that the toxicity of these compounds on other terrestrial organisms (such as microbes, worms, insects and other invertebrates) has not been investigated).

The combination of unlikely introductions of these drugs into the terrestrial environment and the limited toxicity demonstrated in several terrestrial species indicate that significant effects appear unlikely in many of the organisms present in the terrestrial environment.

D. Conclusions

The potential for adverse environments effects due to the use of Romet-30 centers on the aqueous discharges and fish wastes resulting from salmonid culture. Simple dilution in the receiving waters would appear to preclude long-term irreversible environmental effects. The requested action to approve 5-day treatment of salmonids at 50 mg drug (a.i.)/Kg fish body weight does not appear to pose a significant increase in the environmental costs associated with the use of the drug.

A more detailed evaluation of the uses of the drug active ingredients (sulfadimethoxine and ormetoprim) is desirable, however. The following information is absent--
1. Additional information on the physical-chemical properties of sulfadimethoxine and ormetoprim.
   a. Vapor pressure
   b. Dissociation constant
   c. Octanol-water partitioning coefficient
   d. Sorption-desorption isotherms for soils and/or sediments

2. Additional information on the inactivation of sulfadimethoxine and ormetoprim antimicrobial activity:
   a. The probable pathway of degradation of the drug molecules
   b. Processes involved in degradation (e.g., chemical, photochemical, and/or biological)
3. Ecological effects data:
   a. Antimicrobial, antifungal and antiprotozoal spectrum of activity, particularly for non-pathogenic, beneficial bacteria, fungi and protozoa.
   b. Effects on representative invertebrete populations present in water and aquatic sediments.

E. Summary

The requested action to approve the use of Romet-30 at 50 mg drug/Kg fish for 5 days of administration to salmonids for the control of furunculosis does not appear to result in a significant increase in the potential for the manufacture and use of the drug to cause adverse environmental impacts. Date gaps, however, indicate that additional studies are desirable in order to evaluate the environmental fate of these drug molecules and the effects that may occur in the vicinity of salmonid growing facilities as a result of use of Romet-30.

References

Brown, E.E. 1983. World Fish Farming: Cultivation and Economics. 2nd ed., chapter 1 (United States of America). AVI Publishing Company, Inc. Westport, CT. 516 pp.

Joint Subcommittee on Aquaculture. 1983. National Aquaculture Development Plan. Two Volumes. U.S. Department of Interior, Washington, D.C. 67 pp. (vol. 1) and 196 pp. (vol. 2).

Kenaga, E. and C.A.I. Goring. 1980. Relationship between water solubility, soil sorption, octanol-water partitioning, and concentration of chemicals in biota. Pp. 78-115 in Aquatic Toxicology, ASTM Special Technical Publication 707, J.G. Eaton, P.R. Parrish, and A.C. Hendricks (eds.). American Society for Testing and Materials, Philadelphia, PA.

9-28-84
Date

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Attachment

cc: Orig. & Dup., (NADA 123-933)
Office File, HFV-152
Reading Board, HFV-152
Draft:MZeeman:cbm:6/11/84
Draft:Revised:cbm:6/13/84
Final:cbm:6/18/84
Revised:MZeeman:cbm:9/27/84

September 27, 1984

Dr. Maurice Zeeman, HFV-152

Revision of FONSI for Romet-30 for Salmonids (NADA 125-933)

Dr. Charles Haines, HFV-133
Through: John C. Matheson III, Chief
Environmental Staff, HFV-152

Mr. Taylor Madill (HFV-230) has brought to our attention an inconsistency in the final claims that were approved for Romet-30 for salmonids and the FONSI that we prepared and you signed on June 25, 1984. The claim being approved now is for the control (and not the prevention and therapy) of furunculosis in salmonids. A claim for the prevention and therapy of enteric red mouth is also apparently not being approved at this time. Please remember to inform us of such changes in claims as soon as you can in the final approval process. This will allow us to allocate the time and resources necessary to make the changes required in the NEPA documents that will be put on public display.

Accordingly, a revised FONSI has been prepared and is attached. Please sign the original FONSI and ensure that it and the entire EIAR (98 pp.) dated February 22, 1984 are both placed on public display at the Dockets Management Branch along with the final regulation approving this NADA. Second, please sign the salmon copy of the FONSI and ensure that it and the duplicate are placed in the NADA jackets. Third, please sign the HFV-152 copy of the FONSI and return it to us for our files. Finally, please notify the NADA sponsor of the action taken.

Attachment

cc: Orig. & Dup. (NADA 125-933)
Office File (HFV-152)
Reading Board (HFV-152)

MZeeman:cbm:9/27/84

M. Zeeman 9-28-84
9-28-84 HFV-152