Risks of Ziram Use to Federally Threatened California Red-legged Frog
(*Rana aurora draytonii*)

Pesticide Effects Determination

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1. Executive Summary

The purpose of this assessment is to evaluate potential direct and indirect effects on the California red-legged frog *(Rana aurora draytonii*) (CRLF) arising from FIFRA regulatory actions regarding use of Ziram on agricultural sites. In addition, this assessment evaluates whether these actions can be expected to result in modification of the species' designated critical habitat. This assessment was completed in accordance with the U.S. Fish and Wildlife Service (USFWS) and National Marine Fisheries Service (NMFS) *Endangered Species Consultation Handbook* (USFWS/NMFS, 1998 and procedures outlined in the Agency's Overview Document (U.S. EPA, 2004).

The CRLF was listed as a threatened species by USFWS in 1996. The species is endemic to California and Baja California (Mexico) and inhabits both coastal and interior mountain ranges. A total of 243 streams or drainages are believed to be currently occupied by the species, with the greatest numbers in Monterey, San Luis Obispo, and Santa Barbara counties (USFWS, 1996) in California.

Zinc-bis(dimethyldithiocarbamate) (Ziram) is a fungicide that is registered for use on variety of fruits and vegetables. The mode of action of the dimethyldithiocarbamate fungicides is apparently complex, probably involving multiple sites. It has been suggested that they may act by interfering with metal enzyme catalysts.

The following uses are labeled in California and are considered as part of the federal action evaluated in this assessment:

- Almonds
- Apples/pears
- Apricots
- Cherries
- Grapes (including wine grapes)
- Peaches/Nectarines
- Pecans

All uses registered in California are summarized in Table 7.

Fate Properties and Exposure Routes

Ziram (Zinc bis(dimethyldithiocarbamate)) is slightly to moderately persistent in the environment with exposure to the degradate Thiram (tetramethylthiram disulfide) extending the duration of exposure. Generally Ziram degrades rapidly via hydrolysis, photodegradation; degradation is somewhat slower in aerobic soil and slower still in anaerobic soil and water. Half-lives are generally from a few days to a few weeks in soil and water; field studies show some residues may persist for months after application.

Thiram, a major degradate of Ziram, is often formed rapidly and is also of toxicological concern. Thiram is also a registered pesticide with agricultural and outdoor uses. Thiram degrades by similar pathways as Ziram but hydrolysis, aerobic metabolism, and anaerobic metabolism tend to be somewhat slower than for Ziram (compare Table 6 with Table 5).

Laboratory and field data suggest that Ziram and its degradate Thiram are not particularly mobile or volatile compounds, and neither leaching nor volatility are expected to play an important role in the dissipation of Ziram.

Overall, in the field, Ziram, although it typically degrades substantially within a few days after application, appears to sometimes take weeks to months to completely degrade to residues presumed to not be of toxicological concern. Thiram exhibits a similar degradation pattern but is somewhat more persistent and is the major degradate observed in the environment. Thiram is the major degradate of toxicological concern and is the only degradate included in this risk assessment. Chronic exposure is primarily to Thiram. No significant monitoring data for Ziram or its degradate Thiram are available to further characterize exposure to Ziram residues

Since CRLFs exist within aquatic and terrestrial habitats, exposure of the CRLF, its prey and its habitats to Ziram are assessed separately for the two habitats. Tier-II aquatic exposure models are used to estimate high-end exposures of Ziram in aquatic habitats resulting from runoff and spray drift from different uses. Peak model-estimated environmental concentrations resulting from different Ziram uses range from 9.89µg/L for cherries to 43.70µg/L for wine grapes.[1]

To estimate Ziram exposures to the terrestrial-phase CRLF, and its potential prey resulting from uses involving Ziram applications, the T-REX model is used for: foliar. AgDRIFT and AGDISP models are also used to estimate deposition of Ziram on terrestrial and aquatic habitats from spray drift. The T-HERPS model is used to allow for further characterization of dietary exposures of terrestrial-phase CRLFs relative to birds.

The effects determination assessment endpoints for the CRLF include direct toxic effects on the survival, reproduction, and growth of the CRLF itself, as well as indirect effects, such as reduction of the prey base or modification of its habitat. Direct effects to the CRLF in the aquatic habitat are based on toxicity information for freshwater fish, which are generally used as a surrogate for aquatic-phase amphibians. In the terrestrial habitat, direct effects are based on toxicity information for birds, which are used as a surrogate for terrestrial-phase amphibians. Given that the CRLF's prey items and designated critical habitat requirements in the aquatic habitat are dependant on the availability of

[1] EECs are the average one-in-ten year return frequency concentrations based upon multiple PRZM-EXAMS model runs with different sets of application dates for each crop (but only one distinct, representative scenario was utilized in modeling of each crop use site – See section 3.2 for details). The nature of the modeling setup means that the estimates for Ziram and Thiram or not additive, exposure could only occur to either of, but not both, of these compounds at the levels estimated.

freshwater aquatic invertebrates and aquatic plants, toxicity information for these taxonomic groups is also discussed. In the terrestrial habitat, indirect effects due to depletion of prey are assessed by considering effects to terrestrial insects, small terrestrial mammals, and frogs. Indirect effects due to modification of the terrestrial habitat are characterized by available data for terrestrial monocots and dicots.

Risk quotients (RQs) are derived as quantitative estimates of potential high-end risk. Acute and chronic RQs are compared to the Agency's levels of concern (LOCs) to identify instances where Ziram use within the action area has the potential to adversely affect the CRLF and its designated critical habitat via direct toxicity or indirectly based on direct effects to its food supply (i.e., freshwater invertebrates, algae, fish, frogs, terrestrial invertebrates, and mammals) or habitat (i.e., aquatic plants and terrestrial upland and riparian vegetation). When RQs for a particular type of effect are below LOCs, the pesticide is determined to have "no effect" on the subject species. Where RQs exceed LOCs, a potential to cause adverse effects is identified, leading to a conclusion of "may affect." If a determination is made that use of Ziram use within the action area "may affect" the CRLF and its designated critical habitat, additional information is considered to refine the potential for exposure and effects, and the best available information is used to distinguish those actions that "may affect, but are not likely to adversely affect" (NLAA) from those actions that are "likely to adversely affect" (LAA) the CRLF and its critical habitat.

Based on the best available information, the Agency makes a May Affect, Likely to Adversely Affect determination for the CRLF from the use of Ziram. Additionally, the Agency has determined that there is the potential for modification of CRLF designated critical habitat from the use of Ziram. All aquatic-phase modeled acute exposures for Ziram resulted in LOCs exceeding the endangered species thresholds for direct effects using the fish as a surrogate for the CRLF. Therefore, there is a "may affect" for Ziram based on those estimates

Using the probit analysis for Ziram as a refinement, the results indicate there is the potential for an individual effect for all use scenarios. Probit analysis indicates a chance of individual effects ranging about 1 in 1 for almonds with an aerial application, apricots with a ground application, apricots with an aerial application, nectarines with a ground application, nectarines with an aerial application, peaches with a ground application, peaches with an aerial application, pecans wine grapes with a ground application and wine grapes with an aerial application to 1.39 for cherries with an aerial application.

In addition to the probit refinement, the EIIS Incident database was reviewed for aquatic incidents. However, no aquatic incidents were reported.

Additional support for the "LAA" determination is based on acute Thiram results. Probit analysis indicates a chance of individual effects ranging about 1 in 1.00 for wine grapes with an aerial application to 4.12 for pears.
All terrestrial-phase modeled acute exposures for Ziram resulted in LOCs exceeding the endangered species thresholds for direct effects using the bird as a surrogate for the

CRLF. Therefore, there is a “may affect” for Ziram based on those estimates. An “LAA” determination was based on T-HERPS results, with acute dose-based RQs exceeding the LOC for all use scenarios.

Additional lines of evidence supporting the “LAA” determination include the probit refinement as well as a review of open literature and terrestrial incidents. Using the probit analysis as a refinement, the results indicate there is the potential for an individual effect for all use scenarios supporting to an “LAA” determination. The chance of individual effects for all uses was about 1 in 1.00.

In addition to the probit refinement, a review of open literature resulted in no endpoints that were more sensitive than those in registrant submitted studies. A review of terrestrial incidents in the EIIS database resulted in no reports of terrestrial animal incidents for Ziram and one report of bird mortality for Thiram. However, this Thiram report was classified as unlikely due to no pesticide detected in the birds

The direct effects assessment also included reviewing the chronic effects of Ziram on the CRLF. Due to the persistence of the degradate Thiram, Thiram toxicity values will be used with Ziram exposures for this assessment. All chronic dose-based RQs also exceeded the LOC for all use scenarios for both T-REX and T-HERPS supporting an “LAA” determination.

Indirect acute dietary RQs for the aquatic-phase CRLF also exceeded the Endangered Species LOC=0.05 for all uses for aquatic invertebrates as prey items resulting in a “may affect”. The probit analysis indicates there is a potential chance for individual effect for all uses except cherries (about 1 in 11.3 probit result). The “LAA” determination is based on the probit analysis using the slope=1.99 with significant results ranging from about 1 in 2.14 for wine grapes with a ground application to about 1 in 8.19 for grapes.

The “LAA” determination for indirect acute effects for the CRLF is supported by no aquatic incidents for either Ziram or Thiram reported in the EIIS database. There no Ziram aquatic endpoints more sensitive than the registrant submitted studies available in open literature.

Indirect dietary effects for the terrestrial-phase CRLF exceed the LOC threshold for all use scenarios, including fish/frog, terrestrial invertebrates and small mammals resulting in a “may affect” for modeled species.

Using fish as prey for the CRLF, all modeled acute exposures for Ziram resulted in LOCs exceeding the endangered species thresholds for indirect effects. Therefore, there is a “may affect” for Ziram based on those estimates. An “LAA” determination is based on RQs exceeding the acute risk LOC=0.5 for indirect effects on the CRLF consuming fish for all uses. RQs range from 1.66 for cherries with an aerial application to 5.46 for wine grapes with a ground application. Using the probit analysis as a refinement, the results indicate there is the potential for an individual effect for all use scenarios.

In addition to the probit refinement, a review of the open literature resulted in no endpoints more sensitive than the registrant submitted studies. The EIIS Incident database was reviewed for aquatic incidents with the potential to affect fish as prey items. However, no Ziram aquatic incidents were reported.

In addition to the Ziram analysis, results of Thiram acute analysis also support an “LAA” determination based on acute risk LOC exceedence. RQs range from 4.88 for pears to 41.26 for nectarines.

Using the probit analysis as a refinement, the results indicate there is the potential for an individual effect for all use scenarios. Chance of individual effects range from about 1 in 1 for nectarines, peaches and wine grapes with an aerial application to 4.12 for pears

Other supporting lines of evidence include incident reports. No reported Thiram aquatic animal incidents were reported in the EIIS database.

Indirect dietary effects for the terrestrial-phase CRLF exceed the LOC threshold = 0.05 for all use scenarios for both small and large insects for both Ziram, resulting in a “may affect” determination. There is an “LAA” determination for the effects of Ziram on terrestrial invertebrates consumed by the CRLF based on upper-bound RQs exceeding the LOC=0.5.

Due to the indeterminate endpoint for the Ziram study, additional analysis for lines of evidence included an analysis of the Thiram. The Thiram probit analysis using the default slope=4.5 was used to further refine the determination. The probit analysis indicated significant effects based on the chance of individual effects for all uses. The chance of individual effects ranged from about 1 in to about 1 in 2.56 for almonds and apricots to about 1 in 3.33 for cherries. The results of the Thiram analysis support the “LAA” determination.

A review of ECOTOX resulted in no open literature studies with more sensitive endpoints for Ziram. No terrestrial invertebrate incidents were reported in the EIIS database.

Analysis of indirect dietary effects for the CRLF consuming small mammals resulted in T-REX RQs exceeding the LOC threshold for acute effects. This resulted in a “may affect” determination. The “LAA” determination for indirect effects on the CRLF consuming small mammals is based on acute RQs exceeding the acute risk LOC=0.5 for all uses for Ziram. RQs range from 4.53 for cherries to 12.85 for nectarines/peaches.

A review of the open literature for mammals resulted in no endpoints more sensitive than the registrant submitted studies. The EIIS incident database was reviewed for terrestrial incidents with the potential to affect mammals as prey items. However, no Ziram terrestrial animal incidents were reported.

In addition to acute effects, chronic effects for mammals as prey are also analyzed. RQs exceed the LOC=1 for all uses. Spatial analysis is a refinement for the chronic effect. There is an overlap between areas of the expected adverse affect and where the species is located; therefore, the effect can not be discounted.

Critical Habitat

The determination for critical habitat modification is based on results of an effects analysis for aquatic and terrestrial plants as well as the alteration in habitat due to the effect of reduction in prey. There were no LOCs exceeded for aquatic vascular or non-vascular plants. There was no effect to any terrestrial plants at the highest concentration tested. However, due to the effects of Ziram on prey items there is a 'habitat modification' determination. Based on probit results for acute effects there is a significant chance of individual effects for fish and aquatic invertebrates and therefore, a "habitat modification" determination for aquatic-phase PCEs. Also, based on probit results for acute effects there is a significant chance of individual effects for frogs, terrestrial invertebrates, and small mammals and therefore, a "habitat modification" determination for terrestrial-phase PCEs.

A summary of the risk conclusions and effects determinations for the CRLF and its critical habitat is presented in Table 1 and in Table 2. Use-specific determinations for direct and indirect effects to the CRLF are provided in Table 3 and Table 4. Further information on the results of the effects determination is included as part of the Risk Description in Section 5.2. Given the LAA determination for the CRLF and potential modification of designated critical habitat, a description of the baseline status and cumulative effects for the CRLF is provided in Attachment 2.

Table 1. Effects Determination Summary for Ziram Use and the CRLF**.**

Assessment Endpoint	Effects Determination [1]	Basis for Determination
Survival, growth, and/or reproduction of CRLF individuals	LAA[1]	**Potential for Direct Effects** ***Aquatic-phase (Eggs, Larvae, and Adults)***: For acute aquatic-phase direct effects, the May affect is based on all modeled uses RQs (almonds, apples/pears, apricots, cherries, grapes, nectarines, peaches, pecans, wine grapes) exceeding the LOC using the fish as a surrogate for the CRLF. There was an LAA determination based on the lines of evidence including the probit analysis to calculate the probability of individual effects.

Table 1. Effects Determination Summary for Ziram Use and the CRLF**.**

Assessment Endpoint	Effects Determination [1]	Basis for Determination
		Terrestrial-phase (Juveniles and Adults): For acute and chronic terrestrial-phase direct effects, the "may affect" determination is based on all acute and chronic modeled uses T-Rex RQs exceeding the LOC. T-HERPS acute dose-based RQs exceeded the LOC for all modeled uses (almonds, apples/pears, apricots, cherries, grapes, peaches and pecans). The LAA determination for acute direct effects using the bird as a surrogate resulted from lines of evidence for T-HERPS RQs exceeding the LOC for all use scenarios and the probability analysis estimating the chance of individual effects.
		A refinement following the "May Affect" determination for chronic effects using T-HERPS dose-based RQs also exceeded the LOC for all modeled uses (almonds, apples/pears, apricots, cherries, grapes, peaches and pecans) for chronic effects.
		Potential for Indirect Effects
		Aquatic prey items, aquatic habitat, cover and/or primary productivity *Aquatic Prey:* There was a "may affect" determination for aquatic invertebrates based on LOC exceedences for all use scenarios for acute effects to the CRLF. The "LAA" determination was based on the lines of evidence including probit analysis indicating significant effects for individuals for almonds, apples, apricots, grapes, nectarines, pears, peaches, pecans and wine grapes for both Ziram and the degradate Thiram.
		Aquatic habitat: The indirect habitat effect of Ziram on the CRLF is based on an analysis of the acute effects of aquatic invertebrates, there was a "may affect" based on the LOC exceedence for all use scenarios. There was a significant effect indicated by the probit analysis for all use scenarios for uses except cherries.
		Terrestrial prey items, riparian habitat The RQs for all use scenarios exceeded the LOC threshold (0.05) for terrestrial invertebrates for Ziram. The LAA determination is based on lines-of-evidence including a significant individual effect for all use scenarios for Thiram.
		Acute RQs for fish as prey items exceed LOCS for all uses. There was an "LAA" determination using the Probit analysis as a refinement based on significant chance of individual effects for all uses.
		The T-REX acute and chronic analysis for mammals as prey resulted in a "may affect" Lines of evidence for acute effects on mammals as prey items include RQs exceeding the acute risk LOC=0.5 and the probit analysis resulting in significant chance of individual effects resulting in an LAA determination.

Table 1. Effects Determination Summary for Ziram Use and the CRLF**.**

Assessment Endpoint	Effects Determination [1]	Basis for Determination
		The T-REX analysis for chronic effects of mammals as prey items resulted in RQs exceeding the LOC for all uses. There is an overlap between areas of the expected adverse affect and where the species is located; therefore, the effect can not be discounted.
		Riparian Habitat Effects to riparian habitat were based on alteration in habitat due to reduction in prey. There is a significant reduction in prey for frog using the fish as a surrogate due to RQs exceeding the acute risk LOC = 0.5 for all uses.
		For acute effects, there is a significant reduction in prey for mammals due to RQs exceeding the acute risk LOC = 0.5 for all uses for both Ziram and Thiram..

[1] May affect, likely to adversely affect (LAA)

Table 2. Effects Determination Summary for Ziram Use and CRLF Critical Habitat Impact Analysis.

Assessment Endpoint	Effects Determination [1]	Basis for Determination
Modification of aquatic-phase PCE	Habitat Modification[1]	Aquatic PCEs are based on the effect of Ziram on aquatic and terrestrial plants, as well as the effects on fish and aquatic invertebrates consumed by the CRLF. Although there were no effects on aquatic or terrestrial plants, there were effects on fish and aquatic invertebrates consumed by the CRLF. Based on potential effects for both fish and aquatic invertebrates based on LOC exceedence and the chance of individual effects for all uses there is a habitat modification determination for Ziram.
Modification of terrestrial-phase PCE		Terrestrial PCEs are based on the effect of Ziram on terrestrial plants, as well as the effects on prey consumed by the CRL.F. Although there were no effects on terrestrial plants, there were effects on terrestrial invertebrates, mammals and frogs consumed by the CRLF. Ziram RQs for the bee exceeded the LOC (0.05) for all use scenarios. The more sensitive Thiram RQs for the honey bee exceeded the LOC (0.05) for all use scenarios. The probit analysis indicated significant chance of individual effects for all uses.
		T-REX RQs exceeded the LOC for all use scenarios for acute and chronic dietary effects for small mammals. In addition the probit analysis indicated significant effects for all uses for both Ziram and Thiram.
		Using the bird as a surrogate for frogs, all modeled uses resulted in T-REX RQs exceeding the LOC=0.1. Lines of evidence, including probit analysis indicated an "LAA" determination based on significant chance of individual effect for all uses.

[1] Habitat Modification

Table 3. Ziram Use-specific Direct Effects Determinations1 for the CRLF.

Use(s)	Aquatic Habitat		Terrestrial Habitat	
	Acute	Chronic	Acute	Chronic
Almonds	LAA	NE	LAA	LAA
Apples/pears	LAA	NE	LAA	LAA
Apricots	LAA	NE	LAA	LAA
Cherries	LAA	NE	LAA	LAA
Grapes	LAA	NE	LAA	LAA
Pecans	LAA	NE	LAA	LAA
Peaches/Nectarines	LAA	NE	LAA	LAA
Wine Grapes	LAA	NE	LAA	LAA

[1] NE = No effect; NLAA = May affect, but not likely to adversely affect; LAA = Likely to adversely affect

Table 4. Ziram Use-specific Indirect Effects Determinations1 Based on Effects to Prey

Use(s)	Algae	Aquatic Invertebrates		Terrestrial Invertebrates (Acute)	Aquatic-phase frogs and fish		Terrestrial-phase frogs		Small Mammals	
		Acute	Chronic		Acute	Chronic	Acute	Chronic	Acute	Chronic
Almonds	NE	LAA	NE	LAA	LAA	NE	LAA	LAA	LAA	LAA
Apples/pears	NE	LAA	NE	LAA	LAA	NE	LAA	LAA	LAA	LAA
Apricots	NE	LAA	NE	LAA	LAA	NE	LAA	LAA	LAA	LAA
Cherries	NE	LAA	NE	LAA	LAA	NE	LAA	LAA	LAA	LAA
Grapes	NE	LAA	NE	LAA	LAA	NE	LAA	LAA	LAA	LAA
Pecans	NE	LAA	NE	LAA	LAA	NE	LAA	LAA	LAA	LAA
Peaches/Nectarines	NE	LAA	NE	LAA	LAA	NE	LAA	LAA	LAA	LAA
Wine Grapes	NE	LAA	NE	LAA	LAA	NE	LAA	LAA	LAA	LAA

[1] NE = No effect; NLAA = May affect, not likely to adversely affect; LAA = Likely to adversely affect

Based on the conclusions of this assessment, a formal consultation with the U. S. Fish and Wildlife Service under Section 7 of the Endangered Species Act should be initiated to seek concurrence with the LAA determinations and to determine whether there are reasonable and prudent alternatives and/or measures to reduce and/or eliminate potential incidental take.

When evaluating the significance of this risk assessment's direct/indirect and adverse habitat modification effects determinations, it is important to note that pesticide exposures and predicted risks to the species and its resources (i.e., food and habitat) are not expected to be uniform across the action area. In fact, given the assumptions of drift and downstream transport (i.e., attenuation with distance), pesticide exposure and associated risks to the species and its resources are expected to decrease with increasing distance away from the treated field or site of application. Evaluation of the implication of this non-uniform distribution of risk to the species would require information and assessment techniques that are not currently available. Examples of such information and methodology required for this type of analysis would include the following:

- Enhanced information on the density and distribution of CRLF life stages within specific recovery units and/or designated critical habitat within the action area. This information would allow for quantitative extrapolation of the present risk assessment's predictions of individual effects to the proportion of the population extant within geographical areas where those effects are predicted. Furthermore, such population information would allow for a more comprehensive evaluation of the significance of potential resource impairment to individuals of the species.
- Quantitative information on prey base requirements for individual aquatic- and terrestrial-phase frogs. While existing information provides a preliminary picture of the types of food sources utilized by the frog, it does not establish minimal requirements to sustain healthy individuals at varying life stages. Such information could be used to establish biologically relevant thresholds of effects on the prey base, and ultimately establish geographical limits to those effects. This information could be used together with the density data discussed above to characterize the likelihood of adverse effects to individuals.
- Information on population responses of prey base organisms to the pesticide. Currently, methodologies are limited to predicting exposures and likely levels of direct mortality, growth or reproductive impairment immediately following exposure to the pesticide. The degree to which repeated exposure events and the inherent demographic characteristics of the prey population play into the extent to which prey resources may recover is not predictable. An enhanced understanding of long-term prey responses to pesticide exposure would allow for a more refined determination of the magnitude and duration of resource impairment, and together with the information described above, a more complete prediction of effects to individual frogs and potential modification to critical habitat.

2. Problem Formulation

Problem formulation provides a strategic framework for the risk assessment. By identifying the important components of the problem, it focuses the assessment on the most relevant life history stages, habitat components, chemical properties, exposure routes, and endpoints. The structure of this risk assessment is based on guidance contained in U.S. EPA's *Guidance for Ecological Risk Assessment* (U.S. EPA 1998), the Services' *Endangered Species Consultation Handbook* (USFWS/NMFS 1998) and is consistent with procedures and methodology outlined in the Overview Document (U.S. EPA 2004) and reviewed by the U.S. Fish and Wildlife Service and National Marine Fisheries Service (USFWS/NMFS 2004).

2.1 Purpose

The purpose of this endangered species assessment is to evaluate potential direct and indirect effects on individuals of the federally threatened California red-legged frog *(Rana aurora draytonii*) (CRLF) arising from FIFRA regulatory actions regarding use of Ziram on almonds, apples/pears, apricots, cherries, grapes, peaches/nectarines, pecans and wine grapes. In addition, this assessment evaluates whether use on these crops is expected to result in modification of the species' designated critical habitat. This ecological risk assessment has been prepared consistent with a settlement agreement in the case *Center for Biological Diversity (CBD) vs. EPA et al.* (Case No. 02-1580-JSW(JL)) settlement entered in Federal District Court for the Northern District of California on October 20, 2006.

In this assessment, direct and indirect effects to the CRLF and potential modification to its designated critical habitat are evaluated in accordance with the methods described in the Agency's Overview Document (U.S. EPA 2004). Screening level methods include use of standard models such as PRZM-EXAMS, T-REX, TerrPlant, AgDRIFT, and AGDISP, all of which are described at length in the Overview Document [. Additional refinements include: an analysis of the usage data, a spatial analysis, use of the T-HERPS model and use of the probit analysis Use of such information is consistent with the methodology described in the Overview Document (U.S. EPA 2004), which specifies that "the assessment process may, on a case-by-case basis, incorporate additional methods, models, and lines of evidence that EPA finds technically appropriate for risk management objectives" (Section V, page 31 of U.S. EPA 2004).

In accordance with the Overview Document, provisions of the ESA, and the Services' *Endangered Species Consultation Handbook*, the assessment of effects associated with registrations of Ziram is based on an action area. The action area is the area directly or indirectly affected by the federal action, as indicated by the exceedence of the Agency's Levels of Concern (LOCs). It is acknowledged that the action area for a national-level FIFRA regulatory decision associated with a use of Ziram may potentially involve numerous areas throughout the United States and its Territories. However, for the purposes of this assessment, attention will be focused on relevant sections of the action area including those geographic areas associated with locations of the CRLF and its designated critical habitat within the state of California. As part of the "effects determination," one of the following three conclusions will be reached regarding the potential use of Ziram in accordance with current labels:

- "No effect";
- "May affect, but not likely to adversely affect"; or
- "May affect and likely to adversely affect".

Designated critical habitat identifies specific areas that have the physical and biological features, (known as primary constituent elements or PCEs) essential to the conservation of the listed species. The PCEs for CRLFs are aquatic and upland areas where suitable breeding and non-breeding aquatic habitat is located, interspersed with upland foraging and dispersal habitat.

If the results of initial screening-level assessment methods show no direct or indirect effects (no LOC exceedances) upon individual CRLFs or upon the PCEs of the species' designated critical habitat, a "no effect" determination is made for use of Ziram as it relates to this species and its designated critical habitat. If, however, potential direct or indirect effects to individual CRLFs are anticipated or effects may impact the PCEs of the CRLF's designated critical habitat, a preliminary "may affect" determination is made for the FIFRA regulatory action regarding Ziram.

If a determination is made that use of Ziram within the action area(s) associated with the CRLF "may affect" this species or its designated critical habitat, additional information is considered to refine the potential for exposure and for effects to the CRLF and other taxonomic groups upon which these species depend (e.g., aquatic and terrestrial vertebrates and invertebrates, aquatic plants, riparian vegetation, etc.). Additional information, including spatial analysis (to determine the geographical proximity of CRLF habitat and Ziram use sites) and further evaluation of the potential impact of Ziram on the PCEs is also used to determine whether modification of designated critical habitat may occur. Based on the refined information, the Agency uses the best available information to distinguish those actions that "may affect, but are not likely to adversely affect" from those actions that "may affect and are likely to adversely affect" the CRLF or the PCEs of its designated critical habitat. This information is presented as part of the Risk Characterization in Section 5 of this document.

The Agency believes that the analysis of direct and indirect effects to listed species provides the basis for an analysis of potential effects on the designated critical habitat. Because Ziram is expected to directly impact living organisms within the action area (defined in Section 2.7), critical habitat analysis for Ziram is limited in a practical sense to those PCEs of critical habitat that are biological or that can be reasonably linked to biologically mediated processes (i.e., the biological resource requirements for the listed species associated with the critical habitat or important physical aspects of the habitat that may be reasonably influenced through biological processes). Activities that may modify critical habitat are those that alter the PCEs and appreciably diminish the value of the habitat. Evaluation of actions related to use of Ziram that may alter the PCEs of the

CRLF's critical habitat form the basis of the critical habitat impact analysis. Actions that may affect the CRLF's designated critical habitat have been identified by the Services and are discussed further in Section 2.6.

2.2 Scope

Zinc-bis(dimethyldithiocarbamate) (Ziram) is a fungicide that is registered for use on variety of fruits and vegetables. The major degradate of concern is Thiram (tetramethylthiram disulfide), which is also a registered fungicide. Ziram is a metal (zinc) – thiazole chelate with Thiram being the derivative of toxicological concern when the zinc chelation is broken in Ziram.

Currently, labeled uses of ZIRAM include almonds, apples/ pears, apricots, cherries, grapes, peaches/ nectarines, pecans, and wine grapes.

The end result of the EPA pesticide registration process (*i.e.*, the FIFRA regulatory action) is an approved product label. The label is a legal document that stipulates how and where a given pesticide may be used. Product labels (also known as end-use labels) describe the formulation type (*e.g.*, liquid or granular), acceptable methods of application, approved use sites, and any restrictions on how applications may be conducted. Thus, the use or potential use of Ziram in accordance with the approved product labels for California is "the action" relevant to this ecological risk assessment.

Although current registrations of Ziram allow for use nationwide, this ecological risk assessment and effects determination addresses currently registered uses of Ziram in portions of the action area that are reasonably assumed to be biologically relevant to the CRLF and its designated critical habitat. Further discussion of the action area for the CRLF and its critical habitat is provided in Section 2.7.

Degradates and Exposure Concept

Relatively recently submitted Ziram aerobic soil metabolism studies for three different test soils clearly demonstrate that the primary degradate of concern for which exposure to Ziram may occur is Thiram (MRIDs 46622302; 47005202). In soil, Ziram initially degraded with half-lives that were typically in hours; but residues did persist for much longer than might be expected from this initial degradation period, particularly in field studies. When exposed to sunlight in clear water, both Ziram and Thiram photolyzed rapidly. Under alkaline conditions, Ziram is more slowly transformed over a period of several days. However, the primary degradate Thiram persists significantly longer in acidic waters (hydrolysis half-lives up to about 6 months at pH 5). In all aerobic soil metabolism studies, Thiram was by far the primary degradate. From these lines of evidence from the fate studies, it was determined that this assessment should focus on exposure of the CRLF to Ziram and Thiram. Laboratory and field studies have shown Ziram is substantially converted to the more stable Thiram form in the environment within a few days. The conversion rate is however irregular and, for chronic exposure, modeling of combined residues does not provide higher estimates than a more conservative treatment involving modeling of chronic exposure to Thiram alone.

From the analysis of these fate data, it was determined to access acute exposure modeling efforts focused on parent Ziram. However, the assessment of chronic exposure from Ziram used an assumption of immediate conversion of 100% of the applied Ziram to Thiram. Given the similar toxicity of Thiram and Ziram and overall conservative fate assumptions for modeling Thiram, this methodology provides a conservative (protective) assessment of chronic exposure for chronic risk assessment.

The Agency does not routinely include, in its risk assessments, an evaluation of mixtures of active ingredients, either those mixtures of multiple active ingredients in product formulations or those in the applicator's tank. In the case of the product formulations of active ingredients (that is, a registered product containing more than one active ingredient), each active ingredient is subject to an individual risk assessment for regulatory decision regarding the active ingredient on a particular use site. If effects data are available for a formulated product containing more than one active ingredient, they may be used qualitatively or quantitatively in accordance with the Agency's Overview Document and the Services' Evaluation Memorandum (U.S., EPA 2004; USFWS/NMFS 2004).

Ziram apparently has one registered product that contain multiple active ingredients including Ziram (Polyphase 685) although reference to this product could not be located on the manufacturer's (Troy Chemical Company) website as of this writing; it appears this product was registered only for industrial microbiocide uses). Analysis of the available open literature and acute oral mammalian LD_{50} data for multiple active ingredient products relative to the single active ingredient is provided in Appendix A. The results of this analysis show that an assessment based on the toxicity of the single active ingredient of Ziram and the degradate of concern (Thiram) is appropriate.

2.3 Previous Assessments

The previous RED for Ziram was completed October 31, 2001. Ziram is a dimethyldithiocarbamate fungicide that is acutely very highly toxic and poses high acute and chronic risk to most endangered and non-endangered aquatic organisms, should the compound enter aquatic habitats. The major sites considered in this risk assessment include terrestrial food and non-food uses. Acute terrestrial toxicity (and chronic risk for mammals) in avian and mammalian species may occur from the application of Ziram to foliage or other wildlife food items mainly due to the compound's higher application rates and multiple applications, rather than the compound's toxicity.

A data-call-in generated several toxicity studies to be reviewed for the CRLF assessment. Aquatic toxicity studies were reviewed for acute aquatic invertebrates, chronic fish, and aquatic plants. Terrestrial toxicity studies were reviewed for avian reproduction and terrestrial plants.

2.4 Stressor Source and Distribution

2.4.1 Environmental Fate Properties

Table 5 lists the fate and transport properties of Ziram and Table 6 lists the properties for Thiram, which is a primary degradate of Ziram. Additional details including input values for aquatic exposure modeling are provided in Table 5 and Table 6 (in the "Aquatic Exposure Assessment" section).

Ziram is overall slightly to moderately persistent and has limited leaching potential in the environment). Generally Ziram degrades rapidly via hydrolysis, photodegradation, and to some extent in aerobic soil. Half-lives range from 0.17 to 42 hours under natural degradative processes such as hydrolysis (acidic to neutral pH) and photolysis but hydrolysis is slower (half-lives of a few days to a few weeks) under alkaline conditions, in anaerobic soil metabolism, and can be slower also in aerobic soil. Half-lives are generally from a few days to a few weeks in soil and water; furthermore, field studies show some residues may persist for months after application.

A major degradate of Ziram, Thiram is often formed rapidly and is also of toxicological concern (Thiram, is also a registered pesticide with agricultural and outdoor uses). Thiram degrades by similar pathways to Ziram but hydrolysis, aerobic metabolism, and anaerobic metabolism tend to be somewhat slower than for Ziram. It differs from Ziram in that hydrolysis rates decrease with decreasing pH and are much slower under acidic conditions (half-lives at pH 5 were 68 to 169 days). Thiram hydrolysis, aerobic metabolism, and anaerobic metabolism tend to be somewhat slower than for Ziram (compare Table 6 with Table 5).

In spite of being subject to rapid degradation by some pathways, overall, Ziram appears after field applications to take weeks to months to completely degrade to residues presumed to not be of toxicological concern. This conclusion is supported by the results of field dissipation studies for both Ziram and Thiram (the primary degradate of Ziram). For Ziram, field dissipation half-lives were categorized as bi-phasic (initial half-lives of 5 to 7 days, second phase half-lives of 144 to 206 days). In Thiram field dissipation studies half-lives were 14 to 63 days from topsoil and much slower "second phase" dissipation rates were observed (as with Ziram); source: Thiram RED, 9/30/2004).

Besides Thiram, other detected environmental degradates include dimethyldithiocarbamic acid (DDC), N.N-dimethylformamide (DMF), and N.N-dimethylthioforamide (DMTF). None of these degradates are typically found at more than a few percent of the applied in the environment (although they were sometimes detected at a maximum of greater than 10% of the original amount of parent. Chemical structures are provided in Appendix M.

Ziram has limited soil mobility (K_f 2.9 to 68.1 and K_{oc} 314-3732 in four test soils) The leaching potential is also somewhat mitigated by substantial transformation of Ziram to Thiram and then to secondary degradates during the first several days and weeks after application. Should the chemical reach anaerobic regions in the subsoil, the level of persistence may be increased (half-life in the anaerobic soil metabolism study = 14.1 days); the degradate Thiram is generally less mobile in soil than Ziram (for Thiram, the K_f was 54 to 263 and the K_{oc} 2245 to 12899 in four test soils). Overall, neither leaching nor volatility (both Ziram and Thiram have been calculated to have negligible volatility) are expected to play an important role in the dissipation of Ziram.

Thiram is the major degradate of toxicological concern and is the only degradate included in this risk assessment. Chronic exposure is assumed to be primarily to Thiram.

Exposure Routes and Exposure Estimation

While Ziram can reach surface water by spray drift or runoff (it is relatively highly soluble and does not bind to most soils), it is not likely to persist in water for more than a few hours except possibly under alkaline conditions. Thiram is significantly more persistent in water, especially in acidic to neutral waters where hydrolysis half-lives may be from a few days to several months. Therefore, chronic exposure is likely to be mainly to Thiram. With regards to acute exposure, parent Ziram (and its degradate Thiram) may pose ecological risk to aquatic organisms through pulse dosing, due to the compound's high application rates, moderate persistence in soil (allowing more time for residues to be displaced through runoff), multiple applications and short intervals.

Table 5 Environmental fate data for parent Ziram.

Parameter	Value	Calculation Source and Method*
Chemical name	Ziram	---
PC Code	034805	---
Molecular Weight	307.5	MRID 442284-01.
Solubility (mg/L or ppm)	65	Product chemistry data.
Henry's Law Constant	Non-volatile	Agrochemical handbook, 3rd edition.
pH 5 Hydrolysis half life (days)	0.07 (NU)	MRID 43866701. Not used for model input.
pH 7 Hydrolysis half life (days)	0.74	MRID 43866701.
pH 9 Hydrolysis half life (days)	6.3 (NU)	MRID 43866701. Not used for model input.
Soil Photolysis half life (days)	0.3 (NU)	MRID 43642501.
Aquatic photolysis half life (days) (EXAMS KDP input)	0.43	MRID 44097701. Higher of 2 studies, both at pH 5.
Aerobic soil metabolism half life (days) (PRZM DWRATE input)	4.9	MRIDs 43985801; 46622302; 47005202. 90%ile upper C.I. on mean log linear first order $t_{1/2}$ from four studies.
Aerobic aquatic (water column) metabolism half life	Stable	Guidance: If no aerobic aquatic metabolism data are available and there

Table 5 Environmental fate data for parent Ziram.		
Parameter	**Value**	**Calculation Source and Method***
(days) (EXAMS KBACW input)		is significant hydrolysis, assume that the compound is stable to aquatic metabolism.
Anaerobic soil metabolism half life (days)	14.1	MRID 44228402. Not used in modeling.
Anaerobic aquatic (benthic) metabolism half life (days) (EXAMS KBACS input)	Stable	Guidance: If no anaerobic aquatic metabolism data are available and significant hydrolysis occurs, assume that the compound is stable to aquatic metabolism.
K_d or K_f (ml/g) (PRZM KD input)	5.7 (input value) (Range 2.9-68.1)	MRID 43873501. Lowest non- sand K_f (K_{oc} highly variable between 4 test soils).
K_{oc} (ml/g)	314-3732	MRID 43873501. Not used for model input.

Table 6. Environmental fate data for Thiram (as a Ziram metabolite).		
Chemical name	**Thiram**	**Calculation Source and Method***
PC Code	079801	---
Molecular Weight	240.44	Product chemistry data
Solubility (mg/L or ppm)	16	Product chemistry data
Henry's Law Constant	0	Agrochemical handbook, 3rd edition.
pH 5 Hydrolysis half life (days)	68.50; 169.00	MRIDs 45714101 and 41840601. Not used for model input.
pH 7 Hydrolysis half life (days)	17.9 (measured value of 3.5 days not used for model input)	MRIDs 45714101 and 41840601. Most conservative of values measured in two studies (both classified as supplemental); $t_{1/2}$ =3.5 days in other study.
pH 9 Hydrolysis half life (days)	0.29; 6.90	MRIDs 45714101 and 41840601. Not used for model input
Soil Photolysis half life (days)	0.72	MRID 45724501. Not used for model input.
Aquatic photolysis half life (days) (EXAMS KDP input)	0.36	MRID 41753801.
Aerobic soil metabolism half life (days) (PRZM DWRATE input)	17.2	Over all log linear first order $t_{1/2}$ (Not multiplied by 3 because inference from multiple Ziram studies is that the $t_{1/2}$ is

Table 6. Environmental fate data for Thiram (as a Ziram metabolite).

Chemical name	Thiram	Calculation Source and Method*
		typically shorter than 17 days.
Aerobic aquatic (water column) metabolism half life (days) (EXAMS KBACW input)	2.5	MRID 45243401. 90%ile upper C.I. on mean log linear first order $t_{1/2}$ from two studies.
Anaerobic soil metabolism half life (days)	No valid study.	Not used in modeling.
Anaerobic aquatic (benthic) metabolism half life (days) (EXAMS KBACS input)	129.3	MRID 43628501. Guidance: If only one half-life value is available, use 3x the measured $t_{1/2}$ (43.1 days in this case).
K_d or K_f (ml/g) (PRZM KD input)	54 (model input) (54 to 267 in 4 test soils)	MRID 43787501. Lowest non- sand K_f (K_{oc} highly variable between 4 test soils.)
K_{oc} (ml/g)	2245 to 24526	MRID 43787501. Not used for model input.

* "Guidance" refers to the "Guidance for Selecting Input Parameters in Modeling the Environmental Fate and Transport of Pesticides": Version II dated February 28, 2002; unless otherwise specified. See: http://www.epa.gov/oppefed1/models/water/input_guidance2_28_02.htm

2.4.1 Environmental Transport Mechanisms

Potential transport mechanisms include pesticide surface water runoff, spray drift, and secondary drift of volatilized or soil-bound residues leading to deposition onto nearby or more distant ecosystems. Surface water runoff and spray drift are expected to be the major routes of exposure for Ziram.

In general, deposition of drifting pesticides is expected to be greatest close to the site of application. Computer models of spray drift (AgDRIFT and/or AGDISP) are used to determine potential exposures to aquatic and terrestrial organisms via spray drift.

2.4.2 Mechanism of Action

Ziram, Zinc-bis(dimethyldithiocarbamate), is a fungicide that is registered for use on variety of crops, including almonds, apples/pears, apricots, cherries, grapes, peaches/nectarines, pecans and wine grapes. The mode of action of the dimethyldithiocarbamate fungicides is apparently complex, probably involving multiple sites. It has been suggested that they may act by interfering with metal enzyme catalysts.

2.4.3 Use Characterization

Analysis of labeled use information is the critical first step in evaluating the federal action. The current label for Ziram represents the FIFRA regulatory action; therefore, labeled use and application rates specified on the label form the basis of this assessment. The assessment of use information is critical to the development of the action area and selection of appropriate modeling scenarios and inputs.

Table 7 presents the uses and corresponding application rates and methods of application considered in this assessment.

Table 7. Ziram Uses Assessed for the CRLF

Use (Application Method)	Max. Single Appl. Rate (lb ai/A)	Max. Rate Application per Year[1]
Apples (West)[2] (Aerial)	4.6 lbs a.i./Acre 4X Per Season	18.4 lbs a.i./Acre
Almonds (Aerial and Ground)	6.1 lbs a.i./Acre 4X Per Season	24.4 lbs a.i./Acre
Apricots (Aerial and Ground)	6.1 lbs a.i./Acre 4X Per Season	24.4 lbs a.i./Acre
Cherries California (Aerial and Ground)	3.8 lbs a.i./Acre 4X Per Season	15.2 lbs a.i./Acre
Grapes (West)[2] (Aerial and Ground)	3.0 lbs a.i./Acre	Not Specified
Peaches/Nectarines (Western US for leaf curl)[2] (Aerial and Ground)	7.6 lbs a.i./Acre 6X Per Season	45.6 lbs a.i./Acre
Pecans (Ground)	6.1 lbs a.i./Acre 6X Per Season	36.6 lbs a.i./Acre
Pears (Ground)	4.6 lbs a.i./Acre 4X Per Season	18.4 lbs a.i./Acre
Wine Grapes (Aerial and Ground)	3.0 lbs/a.i. /Acre	Not specified

Table 7. Ziram Uses Assessed for the CRLF

Use (Application Method)	Max. Single Appl. Rate (lb ai/A)	Max. Rate Application per Year[1]
[1] Note all crops have only 1 cycle or growing season per year. [2] Label directions listed are for West denotes crops grown to the west of the Rockies. (No specification is for crops grown anywhere in the US.)		

The Agency's Biological and Economic Analysis Division (BEAD) provides an analysis of both national- and county-level usage information (Kaul and Jones, 2006) using state-level usage data obtained from USDA-NASS[2], Doane (www.doane.com; the full dataset is not provided due to its proprietary nature) and the California's Department of Pesticide Regulation Pesticide Use Reporting (CDPR PUR) database[3]. CDPR PUR is considered a more comprehensive source of usage data than USDA-NASS or EPA proprietary databases, and thus the usage data reported for Ziram by county in this California-specific assessment were generated using CDPR PUR data. Data from CDPR PUR were obtained for every pesticide application made on every use site at the section level (approximately one square mile) of the public land survey system. BEAD summarized these data to the county level by site, pesticide, and unit treated (Appendix B). Calculating county-level usage involved summarizing across all applications made within a section and then across all sections within a county for each use site and for each pesticide. The county level usage data that were calculated include: average annual pounds applied, average annual area treated, and average and maximum application rate across an eight year period. The units of area treated are also provided where available.

A summary of Ziram usage for all California use sites is provided below in Table 8. Almonds have been by far the crop use site with the highest amount of Ziram use; peaches are the second most important use, and nectarines, pears, and grapes also have significant amounts of Ziram applied.

Table 8. Summary of California Department of Pesticide Registration (CDPR) Pesticide Use Reporting (PUR) Data from 1999 to 2006 for Currently Registered Ziram Uses.

Site Name	Average Pounds Applied / Yr.	Avg App Rate	Avg 95th%ile App Rate	Avg 99th%ile App Rate	"Avg." Max. Annual App Rate reported[1]
ALMOND	529,989.48	4.91	6.8	7.87	27.15

[2] United States Depart of Agriculture (USDA), National Agricultural Statistics Service (NASS) Chemical Use Reports provide summary pesticide usage statistics for select agricultural use sites by chemical, crop and state. See http://www.usda.gov/nass/pubs/estindx1.htm#agchem.

[3] The California Department of Pesticide Regulation's Pesticide Use Reporting database provides a census of pesticide applications in the state. See http://www.cdpr.ca.gov/docs/pur/purmain.htm.

Table 8. Summary of California Department of Pesticide Registration (CDPR) Pesticide Use Reporting (PUR) Data from 1999 to 2006 for Currently Registered Ziram Uses.

Site Name	Average Pounds Applied / Yr.	Avg App Rate	Avg 95th%ile App Rate	Avg 99th%ile App Rate	"Avg." Max. Annual App Rate reported[1]
APPLE	4,011.19	4.75	7	8.09	8.33
APRICOT	15,128.07	4.73	7.09	11.41	12.25
CHERRY	1,109.30	4.25	6.35	6.57	6.57
GRAPE	43,101.76	2.31	3.21	3.57	7.8
GRAPE, WINE	11,094.77	2.91	3.92	4.44	9.34
NECTARINE	67,623.39	5.28	7.26	9.06	12.15
PEACH	159,286.41	5.1	7.78	9.22	21.68
PEAR	46,003.46	4.83	5.56	6.33	19.41

[1] Average of the maximum annual application rate reported by county over the eight years of reporting included in the table. Some reporting or product end user errors are possible; also some maximum application rates on product labels have been lowered since these data were collected.

Assessed Species

The CRLF was federally listed as a threatened species by USFWS effective June 24, 1996 (USFWS 1996). It is one of two subspecies of the red-legged frog and is the largest native frog in the western United States (USFWS 2002). A brief summary of information regarding CRLF distribution, reproduction, diet, and habitat requirements is provided in Sections 2.5.1 through 2.5.4, respectively. Further information on the status, distribution, and life history of and specific threats to the CRLF is provided in Attachment 1.

Final critical habitat for the CRLF was designated by USFWS on April 13, 2006 (USFWS 2006; 71 FR 19244-19346). Further information on designated critical habitat for the CRLF is provided in Section 2.6.

2.5.1 Distribution

The CRLF is endemic to California and Baja California (Mexico) and historically inhabited 46 counties in California including the Central Valley and both coastal and interior mountain ranges (USFWS 1996). Its range has been reduced by about 70%, and the species currently resides in 22 counties in California (USFWS 1996). The species has an elevation range of near sea level to 1,500 meters (5,200 feet) (Jennings and Hayes 1994); however, nearly all of the known CRLF populations have been documented below 1,050 meters (3,500 feet) (USFWS 2002).

Populations currently exist along the northern California coast, northern Transverse Ranges (USFWS 2002), foothills of the Sierra Nevada (5-6 populations), and in southern

California south of Santa Barbara (two populations) (Fellers 2005a). Relatively larger numbers of CRLFs are located between Marin and Santa Barbara Counties (Jennings and Hayes 1994). A total of 243 streams or drainages are believed to be currently occupied by the species, with the greatest numbers in Monterey, San Luis Obispo, and Santa Barbara counties (USFWS 1996). Occupied drainages or watersheds include all bodies of water that support CRLFs (i.e., streams, creeks, tributaries, associated natural and artificial ponds, and adjacent drainages), and habitats through which CRLFs can move (i.e., riparian vegetation, uplands) (USFWS 2002).

The distribution of CRLFs within California is addressed in this assessment using four categories of location including recovery units, core areas, designated critical habitat, and known occurrences of the CRLF reported in the California Natural Diversity Database (CNDDB) that are not included within core areas and/or designated critical habitat (see **Figure 1**). Recovery units, core areas, and other known occurrences of the CRLF from the CNDDB are described in further detail in this section, and designated critical habitat is addressed in Section 2.6. Recovery units are large areas defined at the watershed level that have similar conservation needs and management strategies. The recovery unit is primarily an administrative designation, and land area within the recovery unit boundary is not exclusively CRLF habitat. Core areas are smaller areas within the recovery units that comprise portions of the species' historic and current range and have been determined by USFWS to be important in the preservation of the species. Designated critical habitat is generally contained within the core areas, although a number of critical habitat units are outside the boundaries of core areas, but within the boundaries of the recovery units. Additional information on CRLF occurrences from the CNDDB is used to cover the current range of the species not included in core areas and/or designated critical habitat, but within the recovery units.

Recovery Units

Eight recovery units have been established by USFWS for the CRLF. These areas are considered essential to the recovery of the species, and the status of the CRLF "may be considered within the smaller scale of the recovery units, as opposed to the statewide range" (USFWS 2002). Recovery units reflect areas with similar conservation needs and population statuses, and therefore, similar recovery goals. The eight units described for the CRLF are delineated by watershed boundaries defined by US Geological Survey hydrologic units and are limited to the elevational maximum for the species of 1,500 m above sea level. The eight recovery units for the CRLF are listed in Table 9 and shown in **Figure 1**.

Core Areas

USFWS has designated 35 core areas across the eight recovery units to focus their recovery efforts for the CRLF (see **Figure 1**). Table 9 summarizes the geographical relationship among recovery units, core areas, and designated critical habitat. The core areas, which are distributed throughout portions of the historic and current range of the species, represent areas that allow for long-term viability of existing populations and reestablishment of populations within historic range. These areas were selected because they: 1) contain existing viable populations; or 2) they contribute to the connectivity of other habitat areas (USFWS 2002). Core area protection and enhancement are vital for maintenance and expansion of the CRLF's distribution and population throughout its range.

For purposes of this assessment, designated critical habitat, currently occupied (post-1985) core areas, and additional known occurrences of the CRLF from the CNDDB are considered. Historically occupied sections of the core areas are not evaluated as part of this assessment because the USFWS Recovery Plan (USFWS 2002) indicates that CRLFs are extirpated from these areas. A summary of currently and historically occupied core areas is provided in Table 9 (currently occupied core areas are bolded). While core areas are considered essential for recovery of the CRLF, core areas are not federally-designated critical habitat, although designated critical habitat is generally contained within these core recovery areas. It should be noted, however, that several critical habitat units are located outside of the core areas, but within the recovery units. The focus of this assessment is currently occupied core areas, designated critical habitat, and other known CNDDB CRLF occurrences within the recovery units. Federally-designated critical habitat for the CRLF is further explained in Section 2.6.

Table 9. California Red-legged Frog Recovery Units with Overlapping Core Areas and Designated Critical Habitat

Recovery Unit [1] (Figure 1)	**Core Areas [2,7] (Figure 1)**	**Critical Habitat Units [3]**	**Currently Occupied (post-1985) [4]**	**Historically Occupied [4]**
Sierra Nevada Foothills and Central Valley (1) (eastern boundary is the 1,500m elevation line)	**Cottonwood Creek (partial) (8)**	--	✓	
	Feather River (1)	BUT-1A-B	✓	
	Yuba River-S. Fork Feather River (2)	YUB-1	✓	
	--	NEV-1[6]		
	Traverse Creek/Middle Fork American River/Rubicon (3)	--	✓	
	Consumnes River (4)	ELD-1	✓	
	S. Fork Calaveras River (5)	--		✓
	Tuolumne River (6)	--		✓
	Piney Creek (7)	--		✓
	East San Francisco Bay (partial)(16)	--	✓	
North Coast Range Foothills and Western Sacramento River Valley (2)	**Cottonwood Creek (8)**	--	✓	
	Putah Creek-Cache Creek (9)	--		✓
	Jameson Canyon – Lower Napa Valley (partial) (15)	--	✓	
	Belvedere Lagoon (partial) (14)	--	✓	

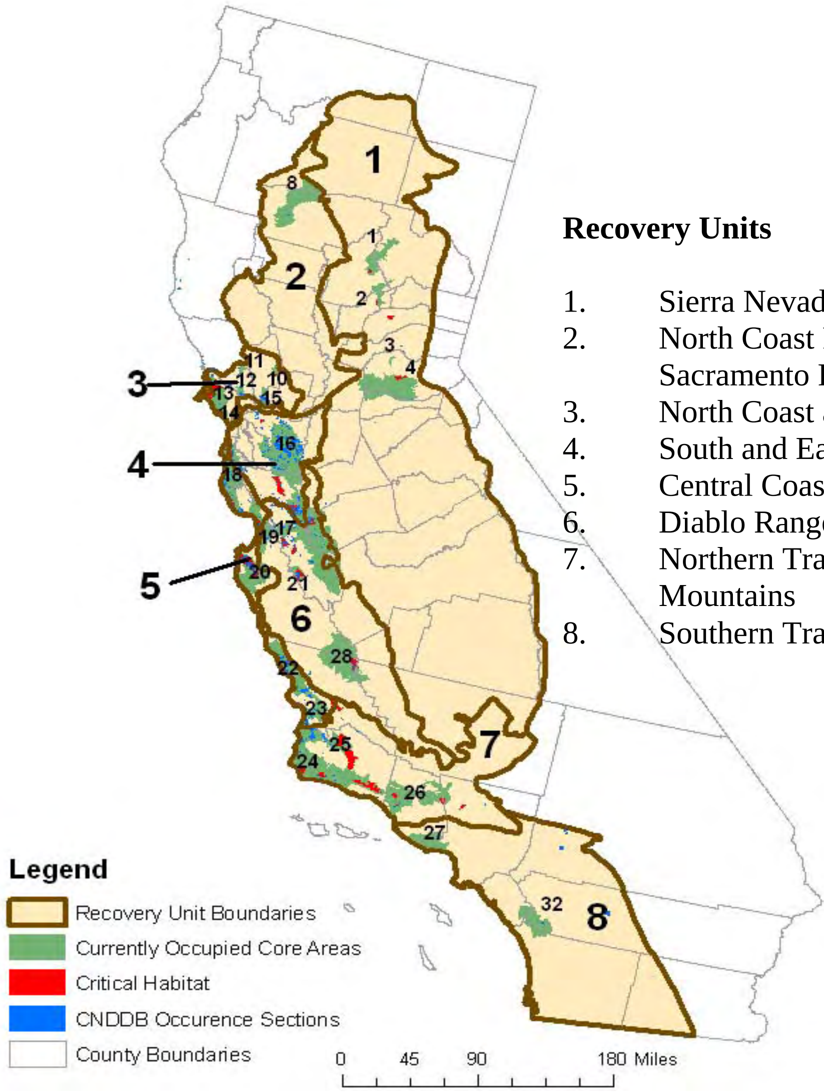
Table 9. California Red-legged Frog Recovery Units with Overlapping Core Areas and Designated Critical Habitat

Recovery Unit [1] (Figure 1)	**Core Areas [2,7] (Figure 1)**	**Critical Habitat Units [3]**	**Currently Occupied (post-1985) [4]**	**Historically Occupied [4]**
	Pt. Reyes Peninsula (partial) (13)	--	✓	
North Coast and North San Francisco Bay (3)	Putah Creek-Cache Creek (partial) (9)	--		✓
	Lake Berryessa Tributaries (10)	NAP-1	✓	
	Upper Sonoma Creek (11)	--	✓	
	Petaluma Creek-Sonoma Creek (12)	--	✓	
	Pt. Reyes Peninsula (13)	MRN-1, MRN-2	✓	
	Belvedere Lagoon (14)	--	✓	
	Jameson Canyon-Lower Napa River (15)	SOL-1	✓	
South and East San Francisco Bay (4)	--	CCS-1A[6]		
	East San Francisco Bay (partial) (16)	ALA-1A, ALA-1B, STC-1B	✓	
	--	STC-1A[6]		
	South San Francisco Bay (partial) (18)	SNM-1A	✓	
Central Coast (5)	**South San Francisco Bay (partial) (18)**	SNM-1A, SNM-2C, SCZ-1	✓	
	Watsonville Slough- Elkhorn Slough (partial) (19)	SCZ-2 [5]	✓	
	Carmel River-Santa Lucia (20)	MNT-2	✓	
	Estero Bay (22)	--	✓	
	--	SLO-8[6]		
	Arroyo Grande Creek (23)	--	✓	
	Santa Maria River-Santa Ynez River (24)	--	✓	
Diablo Range and Salinas Valley (6)	**East San Francisco Bay (partial) (16)**	MER-1A-B, STC-1B	✓	
	--	SNB-1[6], SNB-2[6]		
	Santa Clara Valley (17)	--	✓	
	Watsonville Slough- Elkhorn Slough (partial)(19)	MNT-1	✓	
	Carmel River-Santa Lucia (partial)(20)	--	✓	
	Gablan Range (21)	SNB-3	✓	
	Estrella River (28)	SLO-1A-B	✓	
Northern Transverse Ranges and Tehachapi Mountains (7)	--	SLO-8[6]		
	Santa Maria River-Santa Ynez River (24)	STB-4, STB-5, STB-7	✓	
	Sisquoc River (25)	STB-1, STB-3	✓	
	Ventura River-Santa Clara River (26)	VEN-1, VEN-2, VEN-3	✓	

Table 9. California Red-legged Frog Recovery Units with Overlapping Core Areas and Designated Critical Habitat

Recovery Unit [1] (Figure 1)	**Core Areas [2,7] (Figure 1)**	**Critical Habitat Units [3]**	**Currently Occupied (post-1985) [4]**	**Historically Occupied [4]**
	--	LOS-1[6]		
Southern Transverse and Peninsular Ranges (8)	**Santa Monica Bay-Ventura Coastal Streams (27)**	--	✓	
	San Gabriel Mountain (29)	--		✓
	Forks of the Mojave (30)	--		✓
	Santa Ana Mountain (31)	--		✓
	Santa Rosa Plateau (32)	--	✓	
	San Luis Rey (33)	--		✓
	Sweetwater (34)	--		✓
	Laguna Mountain (35)	--		✓

[1] Recovery units designated by the USFWS (USFWS 2000, pg 49).
[2] Core areas designated by the USFWS (USFWS 2000, pg 51).
[3] Critical habitat units designated by the USFWS on April 13, 2006 (USFWS 2006, 71 FR 19244-19346).
[4] Currently occupied (post-1985) and historically occupied core areas as designated by the USFWS (USFWS 2002, pg 54).
[5] Critical habitat unit where identified threats specifically included pesticides or agricultural runoff (USFWS 2002).
[6] Critical habitat units that are outside of core areas, but within recovery units.
[7] Currently occupied core areas that are included in this effects determination are bolded.



Recovery Units

1. Sierra Nevada Foothills and Central Valley
2. North Coast Range Foothills and Western Sacramento River Valley
3. North Coast and North San Francisco Bay
4. South and East San Francisco Bay
5. Central Coast
6. Diablo Range and Salinas Valley
7. Northern Transverse Ranges and Tehachapi Mountains
8. Southern Transverse and Peninsular Ranges

Figure 1. Recovery Unit, Core Area, Critical Habitat, and Occurrence Designations for CRLF

Core Areas

1. Feather River
2. Yuba River- S. Fork Feather River
3. Traverse Creek/ Middle Fork/ American R. Rubicon
4. Cosumnes River
5. South Fork Calaveras River*
6. Tuolumne River*
7. Piney Creek*
8. Cottonwood Creek
9. Putah Creek – Cache Creek*
10. Lake Berryessa Tributaries
11. Upper Sonoma Creek
12. Petaluma Creek – Sonoma Creek
13. Pt. Reyes Peninsula
14. Belvedere Lagoon
15. Jameson Canyon – Lower Napa River
16. East San Francisco Bay
17. Santa Clara Valley
18. South San Francisco Bay
19. Watsonville Slough-Elkhorn Slough
20. Carmel River – Santa Lucia
21. Gablan Range
22. Estero Bay
23. Arroyo Grange River
24. Santa Maria River – Santa Ynez River
25. Sisquoc River
26. Ventura River – Santa Clara River
27. Santa Monica Bay – Venura Coastal Streams
28. Estrella River
29. San Gabriel Mountain*
30. Forks of the Mojave*
31. Santa Ana Mountain*
32. Santa Rosa Plateau
33. San Luis Ray*
34. Sweetwater*
35. Laguna Mountain*

* Core areas that were historically occupied by the California red-legged frog are not included in the map

Other Known Occurrences from the CNDBB

The CNDDB provides location and natural history information on species found in California. The CNDDB serves as a repository for historical and current species location sightings. Information regarding known occurrences of CRLFs outside of the currently occupied core areas and designated critical habitat is considered in defining the current range of the CRLF. See: http://www.dfg.ca.gov/bdb/html/cnddb_info.html for additional information on the CNDDB.

2.5.2 Reproduction

CRLFs breed primarily in ponds; however, they may also breed in quiescent streams, marshes, and lagoons (Fellers 2005a). According to the Recovery Plan (USFWS 2002), CRLFs breed from November through late April. Peaks in spawning activity vary geographically; Fellers (2005b) reports peak spawning as early as January in parts of coastal central California. Eggs are fertilized as they are being laid. Egg masses are typically attached to emergent vegetation, such as bulrushes (*Scirpus* spp.) and cattails (*Typha* spp.) or roots and twigs, and float on or near the surface of the water (Hayes and Miyamoto 1984). Egg masses contain approximately 2000 to 6000 eggs ranging in size between 2 and 2.8 mm (Jennings and Hayes 1994). Embryos hatch 10 to 14 days after fertilization (Fellers 2005a) depending on water temperature. Egg predation is reported to be infrequent and most mortality is associated with the larval stage (particularly through predation by fish); however, predation on eggs by newts has also been reported (Rathburn 1998). Tadpoles require 11 to 28 weeks to metamorphose into juveniles (terrestrial-phase), typically between May and September (Jennings and Hayes 1994, USFWS 2002); tadpoles have been observed to over-winter (delay metamorphosis until the following year) (Fellers 2005b, USFWS 2002). Males reach sexual maturity at 2 years, and females reach sexual maturity at 3 years of age; adults have been reported to live 8 to 10 years (USFWS 2002).

Figure 2 depicts CRLF annual reproductive timing.

CRLF Reproductive Events by Month

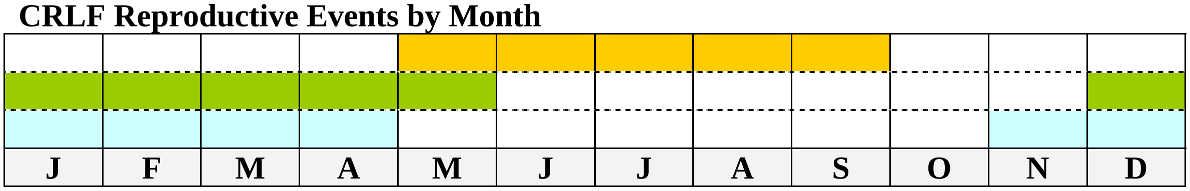


Figure 2. CRLF Reproductive Events by Month.
Light Blue = **Breeding/Egg Masses**
Green = **Tadpoles (except those that over-winter)**
Orange = **Young Juveniles**
Adults and juveniles can be present all year

2.5.3 Diet

Although the diet of CRLF aquatic-phase larvae (tadpoles) has not been studied specifically, it is assumed that their diet is similar to that of other frog species, with the aquatic phase feeding exclusively in water and consuming diatoms, algae, and detritus (USFWS 2002). Tadpoles filter and entrap suspended algae (Seale and Beckvar, 1980) via mouthparts designed for effective grazing of periphyton (Wassersug, 1984, Kupferberg *et al.*; 1994; Kupferberg, 1997; Altig and McDiarmid, 1999).

Juvenile and adult CRLFs forage in aquatic and terrestrial habitats, and their diet differs greatly from that of larvae. The main food source for juvenile aquatic- and terrestrial-phase CRLFs is thought to be aquatic and terrestrial invertebrates found along the shoreline and on the water surface. Hayes and Tennant (1985) report, based on a study examining the gut content of 35 juvenile and adult CRLFs, that the species feeds on as many as 42 different invertebrate taxa, including Arachnida, Amphipoda, Isopoda, Insecta, and Mollusca. The most commonly observed prey species were larval alderflies (*Sialis* cf. *californica*), pillbugs (*Armadilliadrium vulgare*), and water striders (*Gerris* sp). The preferred prey species, however, was the sowbug (Hayes and Tennant, 1985). This study suggests that CRLFs forage primarily above water, although the authors note other data reporting that adults also feed under water, are cannibalistic, and consume fish. For larger CRLFs, over 50% of the prey mass may consists of vertebrates such as mice, frogs, and fish, although aquatic and terrestrial invertebrates were the most numerous food items (Hayes and Tennant 1985). For adults, feeding activity takes place primarily at night; for juveniles feeding occurs during the day and at night (Hayes and Tennant 1985).

2.5.4 Habitat

CRLFs require aquatic habitat for breeding, but also use other habitat types including riparian and upland areas throughout their life cycle. CRLF use of their environment varies; they may complete their entire life cycle in a particular habitat or they may utilize multiple habitat types. Overall, populations are most likely to exist where multiple breeding areas are embedded within varying habitats used for dispersal (USFWS 2002). Generally, CRLFs utilize habitat with perennial or near-perennial water (Jennings et al. 1997). Dense vegetation close to water, shading, and water of moderate depth are habitat features that appear especially important for CRLF (Hayes and Jennings 1988).

Breeding sites include streams, deep pools, backwaters within streams and creeks, ponds, marshes, sag ponds (land depressions between fault zones that have filled with water), dune ponds, and lagoons. Breeding adults have been found near deep (0.7 m) still or slow moving water surrounded by dense vegetation (USFWS 2002); however, the largest number of tadpoles have been found in shallower pools (0.26 – 0.5 m) (Reis, 1999). Data indicate that CRLFs do not frequently inhabit vernal pools, as conditions in these habitats generally are not suitable (Hayes and Jennings 1988).

CRLFs also frequently breed in artificial impoundments such as stock ponds, although additional research is needed to identify habitat requirements within artificial ponds (USFWS 2002). Adult CRLFs use dense, shrubby, or emergent vegetation closely associated with deep-water pools bordered with cattails and dense stands of overhanging vegetation (http://www.fws.gov/endangered/features/rl_frog/rlfrog.html#where).

In general, dispersal and habitat use depends on climatic conditions, habitat suitability, and life stage. Adults rely on riparian vegetation for resting, feeding, and dispersal. The foraging quality of the riparian habitat depends on moisture, composition of the plant community, and presence of pools and backwater aquatic areas for breeding. CRLFs can be found living within streams at distances up to 3 km (2 miles) from their breeding site and have been found up to 30 m (100 feet) from water in dense riparian vegetation for up to 77 days (USFWS 2002).

During dry periods, the CRLF is rarely found far from water, although it will sometimes disperse from its breeding habitat to forage and seek other suitable habitat under downed trees or logs, industrial debris, and agricultural features (UWFWS 2002). According to Jennings and Hayes (1994), CRLFs also use small mammal burrows and moist leaf litter as habitat. In addition, CRLFs may also use large cracks in the bottom of dried ponds as refugia; these cracks may provide moisture for individuals avoiding predation and solar exposure (Alvarez 2000).

2.6 Designated Critical Habitat

In a final rule published on April 13, 2006, 34 separate units of critical habitat were designated for the CRLF by USFWS (USFWS 2006; FR 51 19244-19346). A summary of the 34 critical habitat units relative to USFWS-designated recovery units and core areas (previously discussed in Section 2.5.1) is provided in Table 9.

'Critical habitat' is defined in the ESA as the geographic area occupied by the species at the time of the listing where the physical and biological features necessary for the conservation of the species exist, and there is a need for special management to protect the listed species. It may also include areas outside the occupied area at the time of listing if such areas are 'essential to the conservation of the species.' All designated critical habitat for the CRLF was occupied at the time of listing. Critical habitat receives protection under Section 7 of the ESA through prohibition against destruction or adverse modification with regard to actions carried out, funded, or authorized by a federal Agency. Section 7 requires consultation on federal actions that are likely to result in the destruction or adverse modification of critical habitat.

To be included in a critical habitat designation, the habitat must be 'essential to the conservation of the species.' Critical habitat designations identify, to the extent known using the best scientific and commercial data available, habitat areas that provide essential life cycle needs of the species or areas that contain certain primary constituent elements (PCEs) (as defined in 50 CFR 414.12(b)). PCEs include, but are not limited to, space for individual and population growth and for normal behavior; food, water, air, light, minerals, or other nutritional or physiological requirements; cover or shelter; sites for breeding, reproduction, rearing (or development) of offspring; and habitats that are protected from disturbance or are representative of the historic geographical and ecological distributions of a species. The designated critical habitat areas for the CRLF are considered to have the following PCEs that justify critical habitat designation:

- Breeding aquatic habitat;
- Non-breeding aquatic habitat;
- Upland habitat; and
- Dispersal habitat.

Further description of these habitat types is provided in Attachment 1.

Occupied habitat may be included in the critical habitat only if essential features within the habitat may require special management or protection. Therefore, USFWS does not include areas where existing management is sufficient to conserve the species. Critical habitat is designated outside the geographic area presently occupied by the species only when a designation limited to its present range would be inadequate to ensure the conservation of the species. For the CRLF, all designated critical habitat units contain all four of the PCEs, and were occupied by the CRLF at the time of FR listing notice in April 2006. The FR notice designating critical habitat for the CRLF includes a special rule exempting routine ranching activities associated with livestock ranching from incidental take prohibitions. The purpose of this exemption is to promote the conservation of rangelands, which could be beneficial to the CRLF, and to reduce the rate of conversion to other land uses that are incompatible with CRLF conservation. Please see Attachment 1 for a full explanation on this special rule.

USFWS has established adverse modification standards for designated critical habitat (USFWS 2006). Activities that may destroy or adversely modify critical habitat are those that alter the PCEs and jeopardize the continued existence of the species. Evaluation of actions related to use of Ziram that may alter the PCEs of the CRLF's critical habitat form the basis of the critical habitat impact analysis. According to USFWS (2006), activities that may affect critical habitat and therefore result in adverse effects to the CRLF include, but are not limited to the following:

(1) Significant alteration of water chemistry or temperature to levels beyond the tolerances of the CRLF that result in direct or cumulative adverse effects to individuals and their life-cycles.
(2) Significant increase in sediment deposition within the stream channel or pond or disturbance of upland foraging and dispersal habitat that could result in elimination or reduction of habitat necessary for the growth and reproduction of the CRLF by increasing the sediment deposition to levels that would adversely affect their ability to complete their life cycles.
(3) Significant alteration of channel/pond morphology or geometry that may lead to changes to the hydrologic functioning of the stream or pond and alter the timing, duration, water flows, and levels that would degrade or eliminate the CRLF and/or its habitat. Such an effect could also lead to increased sedimentation and degradation in water quality to levels that are beyond the CRLF's tolerances.
(4) Elimination of upland foraging and/or aestivating habitat or dispersal habitat.
(5) Introduction, spread, or augmentation of non-native aquatic species in stream segments or ponds used by the CRLF.
(6) Alteration or elimination of the CRLF's food sources or prey base (also evaluated as indirect effects to the CRLF).

As previously noted in Section 2.1, the Agency believes that the analysis of direct and indirect effects to listed species provides the basis for an analysis of potential effects on the designated critical habitat. Because Ziram is expected to directly impact living organisms within the action area, critical habitat analysis for Ziram is limited in a practical sense to those PCEs of critical habitat that are biological or that can be reasonably linked to biologically mediated processes.

2.7 Action Area

For listed species assessment purposes, the action area is considered to be the area affected directly or indirectly by the federal action and not merely the immediate area involved in the action (50 CFR 402.02). It is recognized that the overall action area for the national registration of Ziram is likely to encompass considerable portions of the United States based on the large array of agricultural uses. However, the scope of this assessment limits consideration of the overall action area to those portions that may be applicable to the protection of the CRLF and its designated critical habitat within the state of California. The Agency's approach to defining the action area under the provisions of the Overview Document (USEPA 2004) considers the results of the risk assessment process to establish boundaries for that action area with the understanding that exposures below the Agency's defined Levels of Concern (LOCs) constitute a no-effect threshold. For the purposes of this assessment, attention will be focused on the footprint of the action (i.e., the area where pesticide application occurs), plus all areas where offsite transport (i.e., spray drift, downstream dilution, etc.) may result in potential exposure within the state of California that exceeds the Agency's LOCs.

Deriving the geographical extent of this portion of the action area is based on consideration of the types of effects that Ziram may be expected to have on the environment, the exposure levels to Ziram that are associated with those effects, and the best available information concerning the use of Ziram and its fate and transport within the state of California. Specific measures of ecological effect for the CRLF that define the action area include any direct and indirect toxic effect to the CRLF and any potential modification of its critical habitat, including reduction in survival, growth, and fecundity as well as the full suite of sub-lethal effects available in the effects literature. Therefore, the action area extends to a point where environmental exposures are below any measured lethal or sub-lethal effect threshold for any biological entity at the whole organism, organ, tissue, and cellular level of organization. In situations where it is not possible to determine the threshold for an observed effect, the action area is not spatially limited and is assumed to be the entire state of California.

The definition of action area requires a stepwise approach that begins with an understanding of the federal action. The federal action is defined by the currently labeled uses for Ziram. An analysis of labeled uses and review of available product labels was completed. Several of the currently labeled uses are special local needs (SLN) uses or are restricted to specific states and are excluded from this assessment. In addition, a distinction has been made between food use crops and those that are non-food/non-agricultural uses. For those uses relevant to the CRLF, the analysis indicates that, for Ziram, the following agricultural uses are considered as part of the federal action evaluated in this assessment:

- Almonds
- Apples/pears
- Apricots
- Cherries
- Grapes
- Peaches/Nectarines
- Pecans
- Wine Grapes

Following a determination of the assessed uses, an evaluation of the potential “footprint” of Ziram use patterns (i.e., the area where pesticide application occurs) is determined. This “footprint” represents the initial area of concern, based on an analysis of available land cover data for the state of California. The initial area of concern is defined as all land cover types and the stream reaches within the land cover areas that represent the labeled uses described above. A map representing all the land cover types that make up the initial area of concern for Ziram is presented in Figure 3.

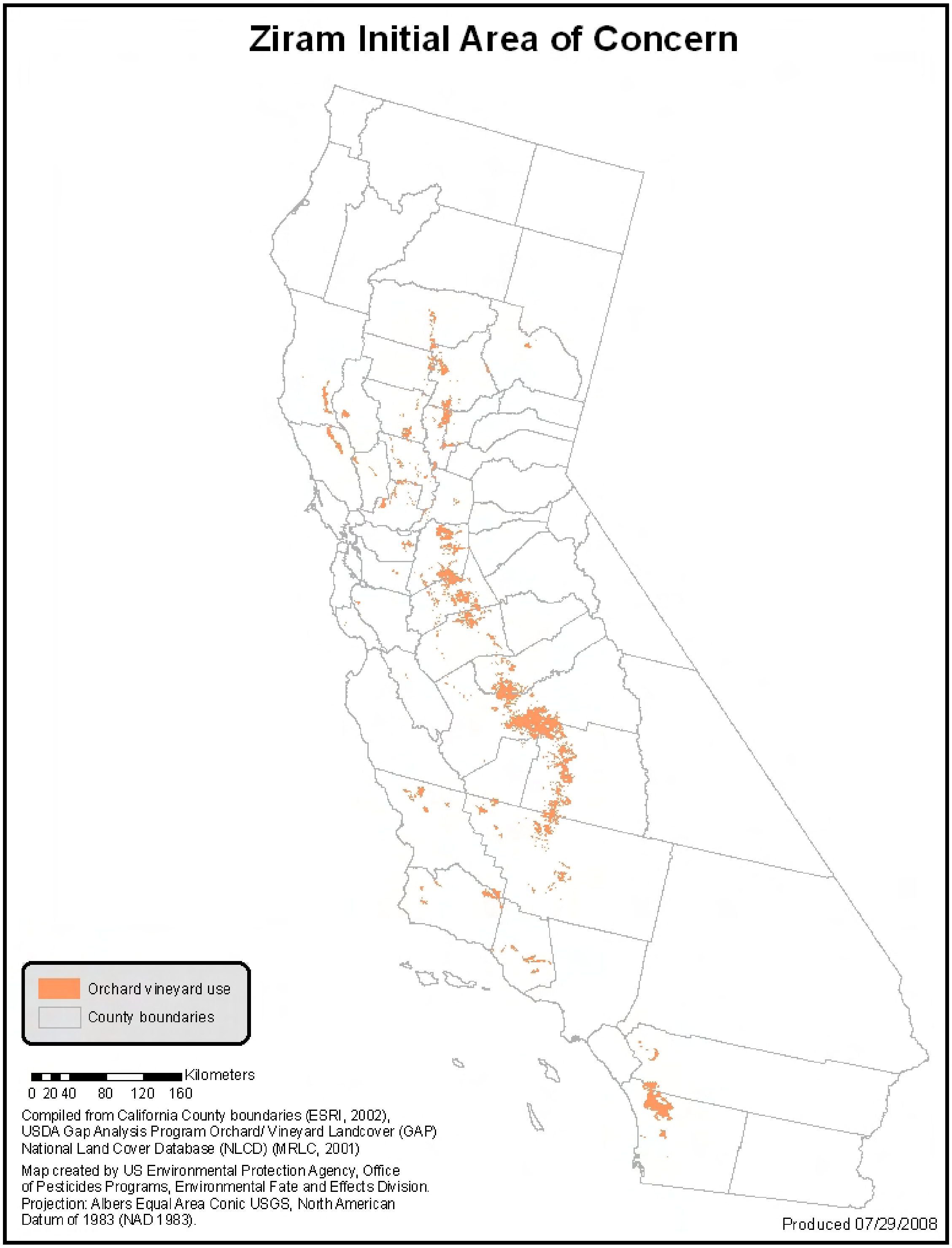


Figure 3. Initial area of concern, or "footprint" of potential use, for Ziram.

Once the initial area of concern is defined, the next step is to define the potential boundaries of the action area by determining the extent of offsite transport via spray drift and runoff where exposure of one or more taxonomic groups to the pesticide exceeds the listed species LOCs.

As previously discussed, the action area is defined by the most sensitive measure of direct and indirect ecological toxic effects including reduction in survival, growth, reproduction, and the entire suite of sub-lethal effects from valid, peer-reviewed studies.

Due to a positive result in a carcinogenicity test (MRID 434042-01 and MRID 457702-01), the spatial extent of the action area (i.e., the boundary where exposures and potential effects are less than the Agency's LOC) for Ziram cannot be determined. Therefore, it is assumed that the action area encompasses the entire state of California, regardless of the spatial extent (i.e., initial area of concern or footprint) of the pesticide use(s).

The geographic extent of reported usage of Ziram in California is outlined in Figure 4 and is summarized by county in Table 10.

Table 10. Summary of CDPR Pesticide Use Reporting by county (annual averages for 1999 to 2006 period).

County	Avg. Annual Pounds Applied	Avg. Annual Area Treated, Acres
Fresno	179277	40764
Butte	90098	17221
Tulare	70002	14033
Madera	68983	14874
Kern	66913	13661
Stanislaus	65181	13787
Merced	59758	13524
Sutter	52435	9122
Glenn	51041	10171
San Joaquin	41655	7623
Yuba	32750	5832
Colusa	19978	4920
Kings	18628	3529
Mendocino	17017	3615
Lake	16286	3211
Tehama	10552	2162
Yolo	7106	1609

Table 10. Summary of CDPR Pesticide Use Reporting by county (annual averages for 1999 to 2006 period).

County	Avg. Annual Pounds Applied	Avg. Annual Area Treated, Acres
Solano	3993	733
Sacramento	3844	659
Contra Costa	1618	251
San Luis Obispo	1180	216
Placer	257	72
San Benito	166	35
Statewide Totals	879032	186709

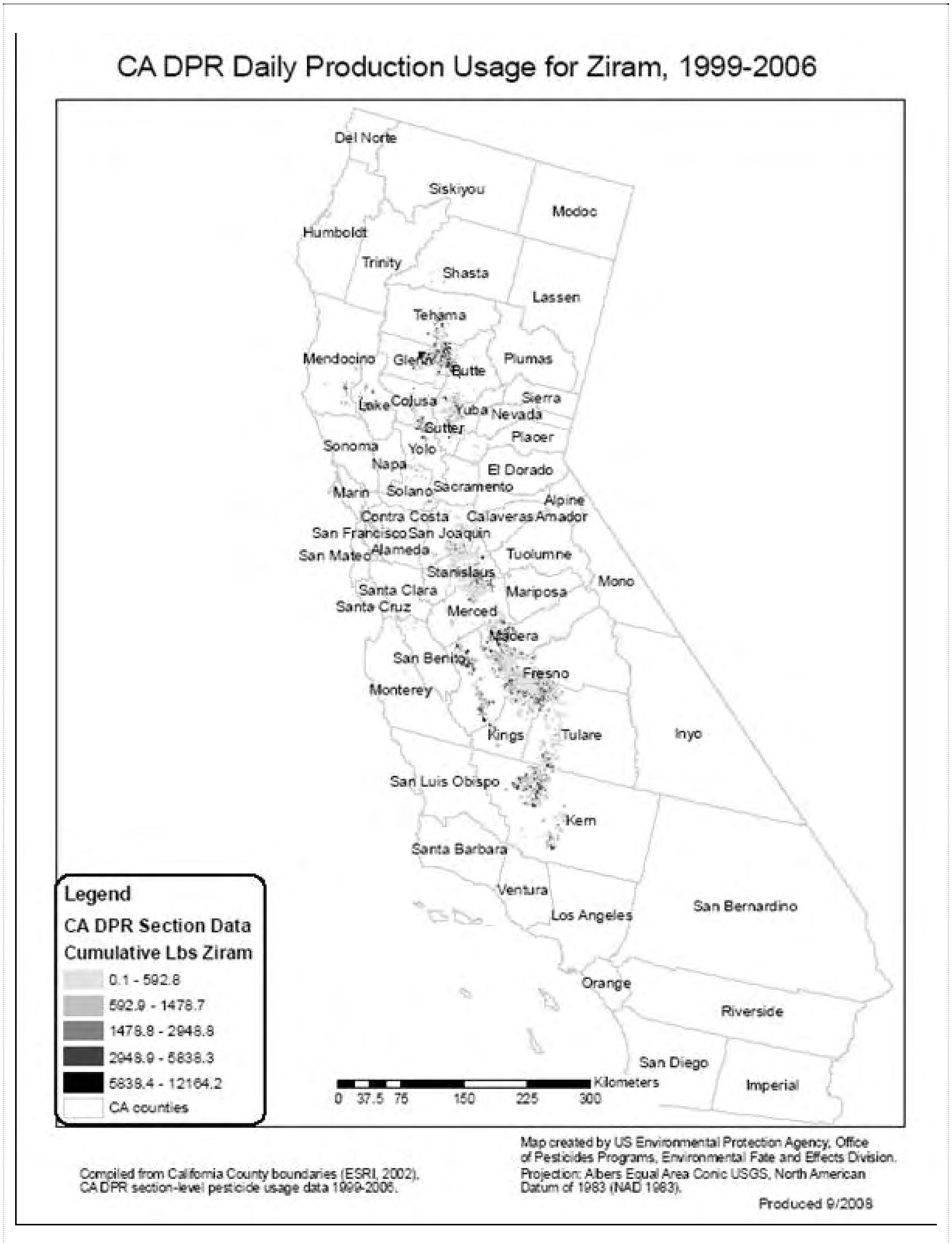


Figure 4. Geographic extent of Ziram usage in California based on 1999 to 2006 data from the PUR database.

2.8 Assessment Endpoints and Measures of Ecological Effect

Assessment endpoints are defined as "explicit expressions of the actual environmental value that is to be protected."[4] Selection of the assessment endpoints is based on valued entities (*e.g.,* CRLF, organisms important in the life cycle of the CRLF, and the PCEs of its designated critical habitat), the ecosystems potentially at risk (*e.g.,* waterbodies, riparian vegetation, and upland and dispersal habitats), the migration pathways of Ziram (*e.g.,* runoff, spray drift.), and the routes by which ecological receptors are exposed to Ziram *(e.g.,* direct contact, *etc.).*

2.8.1. Assessment Endpoints for the CRLF

Assessment endpoints for the CRLF include direct toxic effects on the survival, reproduction, and growth of the CRLF, as well as indirect effects, such as reduction of the prey base or modification of its habitat. In addition, potential modification of critical habitat is assessed by evaluating potential effects to PCEs, which are components of the habitat areas that provide essential life cycle needs of the CRLF. Each assessment endpoint requires one or more "measures of ecological effect," defined as changes in the attributes of an assessment endpoint or changes in a surrogate entity or attribute in response to exposure to a pesticide. Specific measures of ecological effect are generally evaluated based on acute and chronic toxicity information from registrant-submitted guideline tests that are performed on a limited number of organisms. Additional ecological effects data from the open literature are also considered. It should be noted that assessment endpoints are limited to direct and indirect effects associated with survival, growth, and fecundity, and do not include the full suite of sub-lethal effects used to define the action area. According the Overview Document (USEPA 2004), the Agency relies on acute and chronic effects endpoints that are either direct measures of impairment of survival, growth, or fecundity or endpoints for which there is a scientifically robust, peer reviewed relationship that can quantify the impact of the measured effect endpoint on the assessment endpoints of survival, growth, and fecundity.

A complete discussion of all the toxicity data available for this risk assessment, including resulting measures of ecological effect selected for each taxonomic group of concern, is included in Section 4 of this document. A summary of the assessment endpoints and measures of ecological effect selected to characterize potential assessed direct and indirect CRLF risks associated with exposure to Ziram is provided in Table 11.

[4] From U.S. EPA (1992). *Framework for Ecological Risk Assessment.* EPA/630/R-92/001.

Table 11. Assessment Endpoints and Measures of Ecological Effects.

Assessment Endpoint	**Measures of Ecological Effects**[5]
Aquatic-Phase CRLF (Eggs, larvae, juveniles, and adults)[a]	
Direct Effects	
1. Survival, growth, and reproduction of CRLF	1a. Acute Effects: Most sensitive fish acute LC_{50} =0.0097 mg/l for bluegill sunfish (*Lepomis macrochirus*) (MRID 423863-03). Most sensitive cold water fish acute LC50=1.7 mg/l for trout (*Oncorhynchus mykiss*)((MRID 423863-01)
	1b. Chronic Effects: Most sensitive fish early-life stage NOAEC (*Pimephales promelas)* Fathead Minnow (MRID 468931-04) 101 µg/l for post-hatch survival
	No toxicity data for amphibians is available.
Indirect Effects and Critical Habitat Effects	
2. Survival, growth, and reproduction of CRLF individuals via indirect effects on aquatic prey food supply (*i.e.*, fish, freshwater invertebrates, non-vascular plants)	Most sensitive fish (Bluegill, *Lepomis macrochrus*) acute LC_{50} =0.0097 mg/L for MRID 423863-03.
	Most sensitive aquatic invertebrate (*Daphnia*) acute LC_{50} =0.048 mg/L for MRID 423863-05.
	Lemna EC50=370 µg/l. (MRID 468233-02)
	Pseudokirchneriella subcapitata EC50=67 µg/l. (MRID 438339-01)
	Most sensitive fish early-life stage NOAEC for (*Pimephales promelas)* Fathead Minnow (MRID 468931-04) 101 µg/l
	Most sensitive chronic aquatic invertebrate is *Daphnia* 39 µg/l (423863-052)
	No toxicity data for amphibians is available.
3. Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat, cover, food supply, and/or primary productivity (*i.e.*, aquatic plant community)	Aquatic vascular plants: *Lemna* EC50=370 µg/l. (MRID 468233-02)
	Aquatic non-vascular plants: Pseudokirchneriella subcapitata EC50=67 µg/l. (MRID 438339-01)
4. Survival, growth, and reproduction of CRLF individuals via effects to riparian vegetation	Based on the Tier I emergence (MRID 468931-01) and vegetative vigor (MRID 468931-02) toxicity tests, there was no effect to monocots at the highest label rate tested for all modeled uses.

[5] All registrant-submitted and open literature toxicity data reviewed for this assessment are included in Appendix A.

Assessment Endpoint	Measures of Ecological Effects[5]
	Based on the Tier I emergence (MRID 468931-01) and vegetative vigor (MRID468931-02) toxicity tests, there was no effect to dicots at the highest label rate tested for all modeled uses.
Terrestrial-Phase CRLF (Juveniles and adults)	
Direct Effects	
5. Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	(*Colinus virgianus*) Quail LD50=97 mg/kg/bw (MRID 417257-01)
	(*Anas platyrhnchus*) Mallard LC50=5156 mg/kg (MRID 423863-02)
Indirect Effects and Critical Habitat Effects	
6. Survival, growth, and reproduction of CRLF individuals via effects on terrestrial prey (*i.e.*, terrestrial invertebrates, small mammals , and frogs)	(*Colinus virgianus*) Quail LD50=97 mg/kg/bw (MRID 417257-01).
	(*Apis mellifera*) Honey Bee LD50>100 µg/bee (MRID 416679-01)
	(*Rattus norvegicus*) Rat NOAEC=207 mg/kg (MRID 439358-01)
	No data is available for amphibians.
7. Survival, growth, and reproduction of CRLF individuals via indirect effects on habitat (*i.e.*, riparian and upland vegetation)	Based on the Tier I emergence toxicity test (MRID 468931-01) and vegetative vigor (MRID 468931-02), there was no effect to monocots at the highest label rate tested for all modeled uses.
	Based on the Tier I emergence toxicity test (MRID 439358-01) and vegetative vigor (MRID 468931-02), there was no effect to dicots at the highest label rate tested (6.1 lb/A) for all modeled uses.

[a] Adult frogs are no longer in the "aquatic phase" of the amphibian life cycle; however, submerged adult frogs are considered "aquatic" for the purposes of this assessment because exposure pathways in the water are considerably different that exposure pathways on land.
[b] Birds are used as surrogates for terrestrial phase amphibians.

2.8.2 Assessment Endpoints for Designated Critical Habitat

As previously discussed, designated critical habitat is assessed to evaluate actions related to the use of Ziram that may alter the PCEs of the CRLF's critical habitat. PCEs for the CRLF were previously described in Section 2.6. Actions that may modify critical habitat are those that alter the PCEs and jeopardize the continued existence of the CRLF. Therefore, these actions are identified as assessment endpoints. It should be noted that evaluation of PCEs as assessment endpoints is limited to those of a biological nature (i.e., the biological resource requirements for the listed species associated with the critical habitat) and those for which Ziram effects data are available.

Adverse modification to the critical habitat of the CRLF includes, but is not limited to, the following, as specified by USFWS (2006):

1. Alteration of water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs.
2. Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs.
3. Significant increase in sediment deposition within the stream channel or pond or disturbance of upland foraging and dispersal habitat.
4. Significant alteration of channel/pond morphology or geometry.
5. Elimination of upland foraging and/or aestivating habitat, as well as dispersal habitat.
6. Introduction, spread, or augmentation of non-native aquatic species in stream segments or ponds used by the CRLF.
7. Alteration or elimination of the CRLF's food sources or prey base.

Measures of such possible effects by labeled use of Ziram on critical habitat of the CRLF are described in Table 12. Some components of these PCEs are associated with physical abiotic features (e.g., presence and/or depth of a water body, or distance between two sites), which are not expected to be measurably altered by use of pesticides. Assessment endpoints used for the analysis of designated critical habitat are based on the adverse modification standard established by USFWS (2006). -

Table 12. Summary of Assessment Endpoints and Measures of Ecological Effect for Primary Constituent Elements of Designated Critical Habitat.

Assessment Endpoint	Measures of Ecological Effect
Aquatic-Phase CRLF PCEs (Aquatic Breeding Habitat and Aquatic Non-Breeding Habitat)	
Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.	*Lemna gibba* EC50= 370 µg/L (MRID 468233-02) Based on the Tier I emergence toxicity test (MRID 468931-01) and vegetative vigor (MRID 468931-02), there was no effect to monocots at the highest label rate tested for all modeled uses. Based on the Tier I emergence toxicity test (MRID 439358-01) and vegetative vigor (MRID 468931-02), there was no effect to dicots at the highest label rate tested (6.1 lb/A) for all modeled uses.
Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	*Pseudokirchneriella subcapitata* EC50= 67 µg/l (MRID 438339-01)) Based on the Tier I emergence toxicity test (MRID 468931-01) and vegetative vigor (MRID 468931-02), there was no effect to monocots at the highest label rate tested for all modeled uses. Based on the Tier I emergence toxicity test (MRID 439358-01) and vegetative vigor (MRID 468931-02), there was no effect to dicots at the highest label rate tested (6.1 lb/A) for all modeled uses.
	Bluegill *(Leopmis macrochirus)* acute LC50=9.7(MRID 423863-03). Most sensitive NOAEC values for fish or aquatic-phase amphibians and aquatic invertebrates (guideline or ECOTOX) *Daphnia* acute LC50=48 µg/L (MRID 423863-05) (*Pimephales promelas)* Fathead Minnow NOAEC=101 µg/L (MRID 468931-04) *Daphnia* NOAEC=39 µg/L (MRID 423863-05.)
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.*, algae)	*Pseudokirchneriella subcapitata* EC5=67 µg/l (MRID 438339-01)
Terrestrial-Phase CRLF PCEs (Upland Habitat and Dispersal Habitat)	
Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or dripline surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance. Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal Reduction and/or modification of food sources for terrestrial phase juveniles and adults	Based on the Tier I emergence toxicity test (MRID 468931-01) and vegetative vigor (MRID 468931-02), there was no effect to monocots at the highest label rate tested for all modeled uses. Based on the Tier I emergence toxicity test (MRID 439358-01) and vegetative vigor (MRID 468931-02), there was no effect to dicots at the highest label rate tested (6.1 lb/A) for all modeled uses. (*Lepomis macrochirus*) Bluegill LC_{50}=9.7 µg/L (MRID 423863-03) *Daphnia* NOAEC = 39 µg/L (MRID 423863-05.)

Assessment Endpoint	Measures of Ecological Effect
Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	

[a] Physico-chemical water quality parameters such as salinity, pH, and hardness are not evaluated because these processes are not biologically mediated and, therefore, are not relevant to the endpoints included in this assessment.

2.9 Conceptual Model

2.9.1 Risk Hypotheses

Risk hypotheses are specific assumptions about potential adverse effects (*i.e.*, changes in assessment endpoints) and may be based on theory and logic, empirical data, mathematical models, or probability models (U.S. EPA, 1998). For this assessment, the risk is stressor-linked, where the stressor is the release of Ziram to the environment. Note that acute exposure can be to both Ziram and its degradate Thiram, whereas chronic exposure and risk is primarily to Thiram. The following risk hypotheses are presumed for this endangered species assessment:

The labeled use of Ziram within the action area may:

- directly affect the CRLF by causing mortality or by adversely affecting growth or fecundity;
- indirectly affect the CRLF by reducing or changing the composition of food supply;
- indirectly affect the CRLF or modify designated critical habitat by reducing or changing the composition of the aquatic plant community in the ponds and streams comprising the species' current range and designated critical habitat, thus affecting primary productivity and/or cover;
- indirectly affect the CRLF or modify designated critical habitat by reducing or changing the composition of the terrestrial plant community (i.e., riparian habitat) required to maintain acceptable water quality and habitat in the ponds and streams comprising the species' current range and designated critical habitat;
- modify the designated critical habitat of the CRLF by reducing or changing breeding and non-breeding aquatic habitat (via modification of water quality parameters, habitat morphology, and/or sedimentation);
- modify the designated critical habitat of the CRLF by reducing the food supply required for normal growth and viability of juvenile and adult CRLFs;
- modify the designated critical habitat of the CRLF by reducing or changing upland habitat within 200 ft of the edge of the riparian vegetation necessary for shelter, foraging, and predator avoidance.
- modify the designated critical habitat of the CRLF by reducing or changing dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal.

- modify the designated critical habitat of the CRLF by altering chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs.

2.9.2 Diagram

The conceptual model is a graphic representation of the structure of the risk assessment. It specifies the Ziram release mechanisms, biological receptor types, and effects endpoints of potential concern. The conceptual models for aquatic and terrestrial phases of the CRLF are shown in Figure 5 and Figure 6, respectively, and the conceptual models for the aquatic and terrestrial PCE components of critical habitat are shown in Figure 7 and Figure 8, respectively.

Within each figure, exposure routes shown in dashed lines are not quantitatively considered because the contribution of those potential exposure routes to potential risks to the CRLF and modification to designated critical habitat is expected to be negligible. Dotted lines in Figure 5 and Figure 7, effects for aquatic-phase CRLF and aquatic habitat, indicate low concern for potential risks for groundwater or long-range atmospheric transfer. Dotted lines in Figure 6 and Figure 8, effects for terrestrial-phase CRLF and terrestrial habitat, indicate low concern for potential risks for atmospheric transfer.

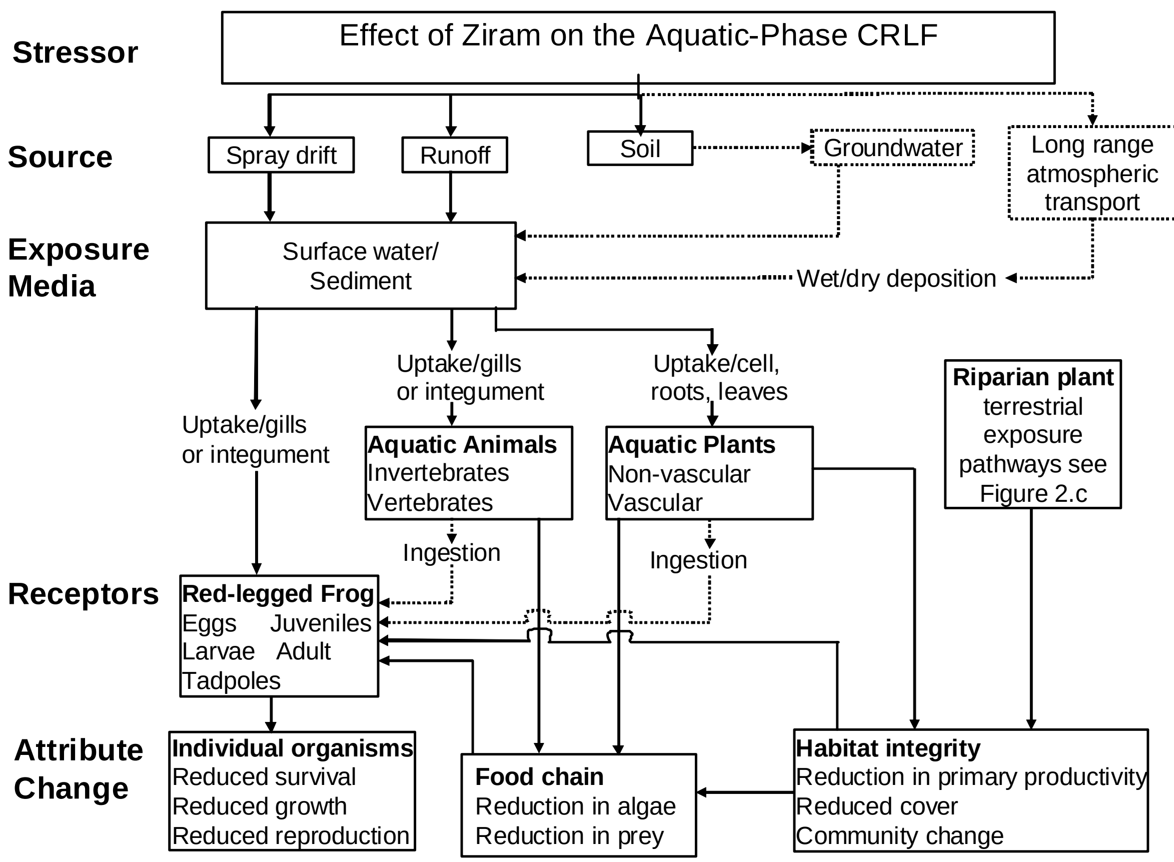


Figure 5. Conceptual Model for Aquatic-Phase of the CRLF.

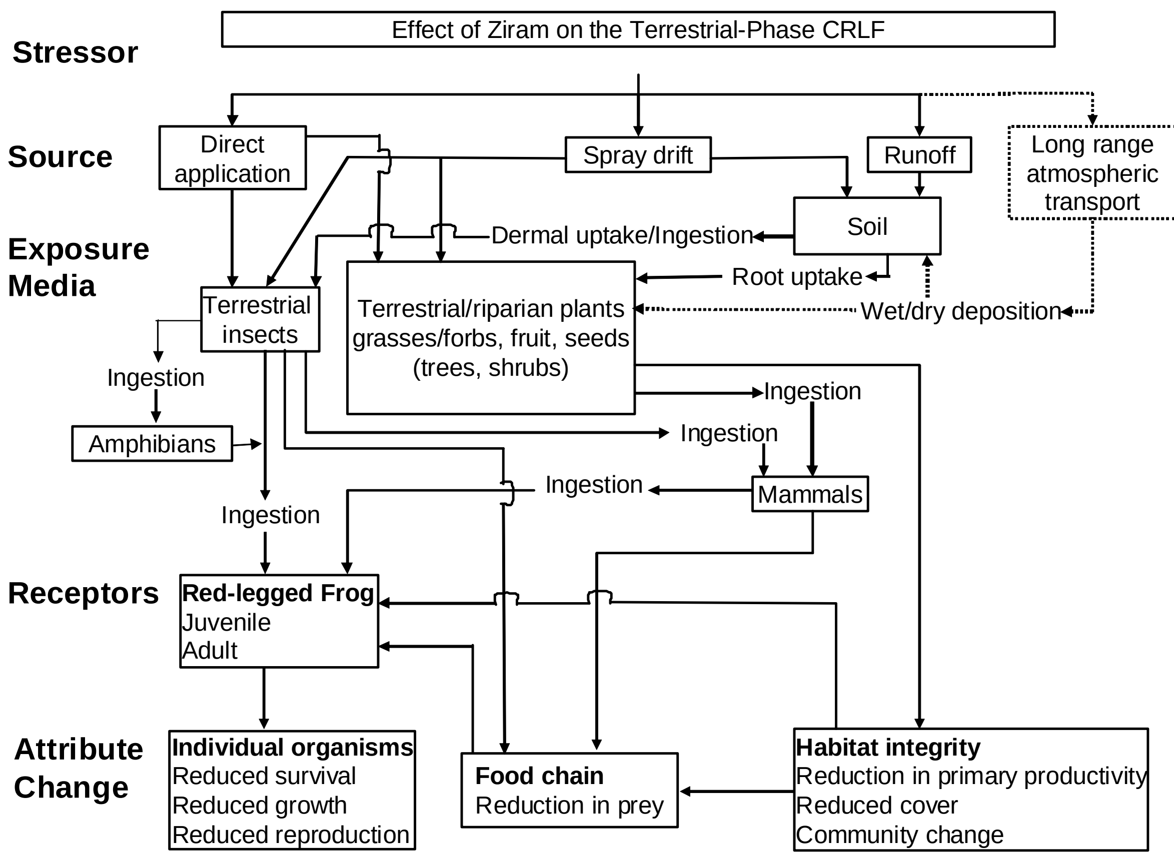


Figure 6. Conceptual Model for Terrestrial-Phase of the CRLF.

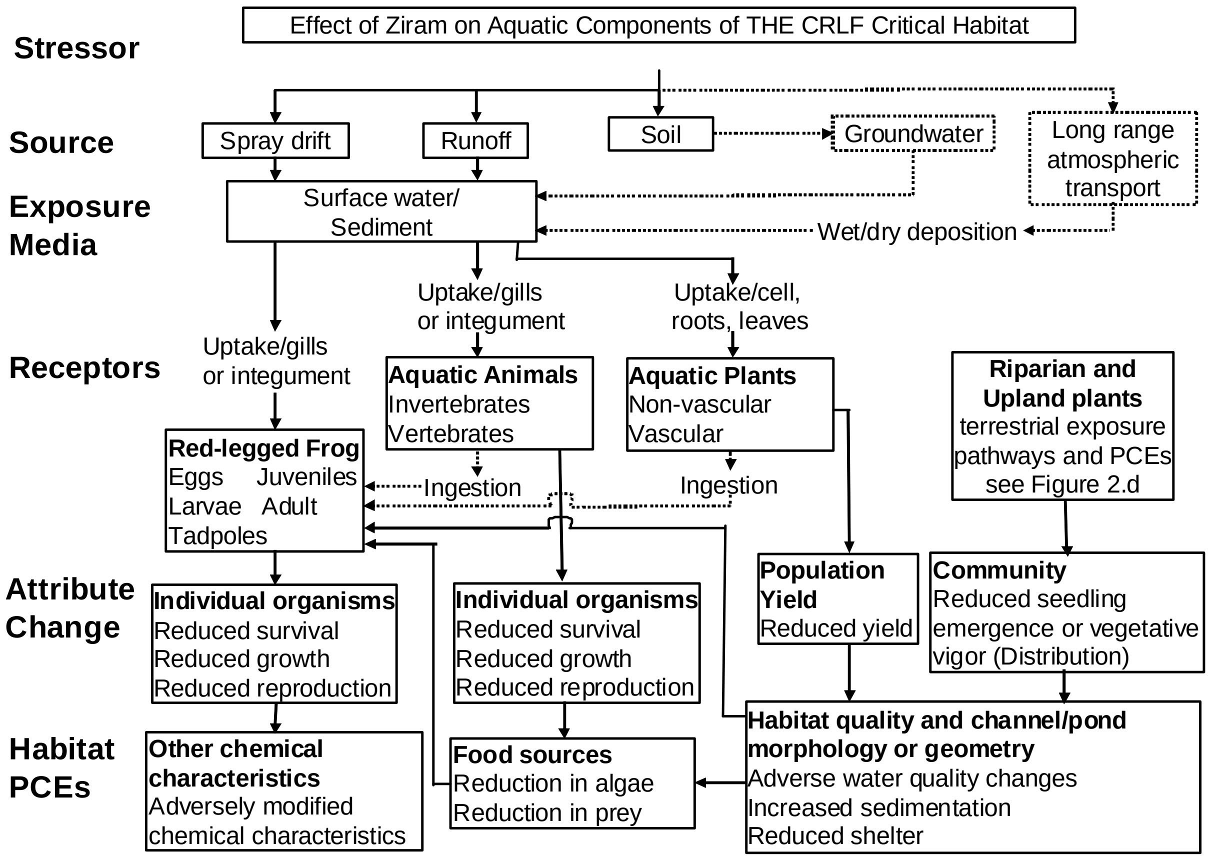


Figure 7. Conceptual Model for Pesticide Effects on Aquatic Component of CRLF Critical Habitat.

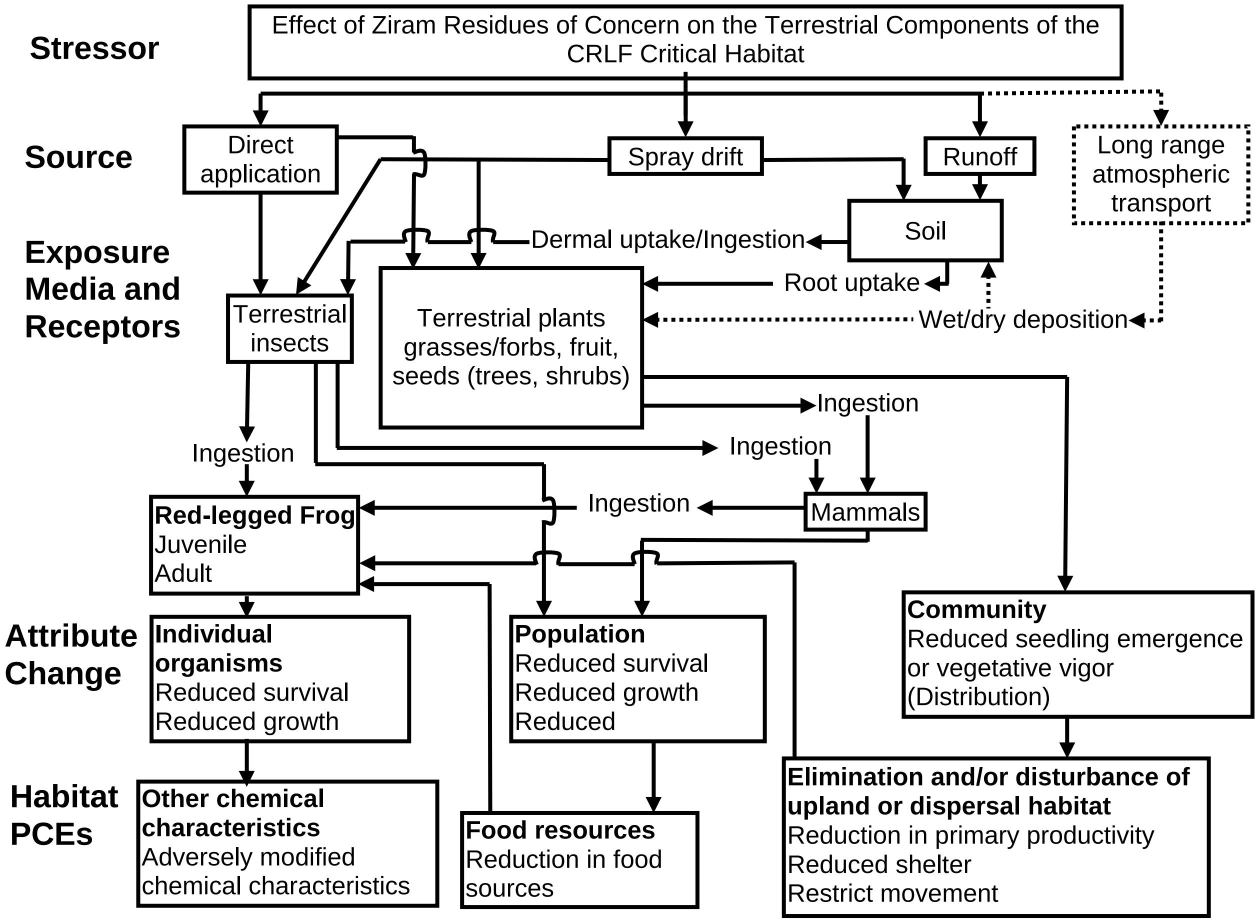


Figure 8. Conceptual Model for Pesticide Effects on Terrestrial Component of CRLF.

2.10 Analysis Plan

In order to address the risk hypothesis, the potential for direct and indirect effects to the CRLF, its prey, and its habitat is estimated. In the following sections, the use, environmental fate, and ecological effects of Ziram are characterized and integrated to assess the risks. Due to the uncertainties involving exposure duration and concentration based on the degradation of Ziram to Thiram, short-term exposure is calculated separately for both Ziram and Thiram using conservative assumptions (results are therefore non-additive), which are present at the same time. Section 4 provides summaries of Ziram toxicity studies used in the assessment. Summaries of all Ziram toxicity studies are found in Appendix C, while Thiram summaries are found in Appendix D. Results of the acute Thiram analysis are found in the risk description as additional lines of evidence.

Long-term exposure is calculated for the more persistent moiety, Thiram. This is accomplished using a risk quotient (ratio of exposure concentration to effects concentration) approach. Although risk is often defined as the likelihood and magnitude of adverse ecological effects, the risk quotient-based approach does not provide a quantitative estimate of likelihood and/or magnitude of an adverse effect. However, as outlined in the Overview Document (U.S. EPA, 2004), the likelihood of effects to individual organisms from particular uses of Ziram is estimated using the probit dose-response slope and either the level of concern (discussed below) or actual calculated risk quotient value.

Aquatic RQ values may be estimated using the acute-to-chronic method for the most sensitive species if no toxicity values are available from acceptable studies. Using both acute and chronic values for Ziram along with acute values for Thiram, the chronic value for Thiram may be estimated as follows:

Chronic endpoint$_{species\ B}$= Acute endpoint species B(Chronic endpoint $_{species\ A}$/acute endpoint $_{species\ A}$). This estimated value will be used in the risk assessment.

2.10.1 Measures to Evaluate the Risk Hypothesis and Conceptual Model

2.10.1.1 Measures of Exposure

The environmental fate properties of Ziram along with available monitoring data indicate that runoff and spray drift are the principle potential transport mechanisms of Ziram to the aquatic and terrestrial habitats of the CRLF. A summary of the fate properties of Ziram and its degradate Thiram is provided in Table 13 and Table 14. In this assessment, transport of Ziram through runoff and spray drift is considered in deriving quantitative estimates of Ziram exposure to CRLF, its prey and its habitats. No information is available on the aerial transport of Ziram, however, it is reported to have negligible volatility which would tend minimize very long range transport.

Measures of exposure are based on aquatic and terrestrial models that predict estimated environmental concentrations (EECs) of Ziram using maximum-labeled application rates and methods of application. The models used to predict aquatic EECs include the Pesticide Root Zone Model coupled with the Exposure Analysis Model System (PRZM/EXAMS). The model used to predict terrestrial EECs on food items is T-REX. The model used to derive EECs relevant to terrestrial and wetland plants is TerrPlant. These models are parameterized using relevant reviewed registrant-submitted environmental fate data.

PRZM (v3.12.2, May 2005) and EXAMS (v2.98.4.6, April 2005) are screening simulation models coupled with the input shell pe5.pl (Aug 2007) to generate daily exposures and 1-in-10 year EECs of Ziram that may occur in surface water bodies adjacent to application sites receiving Ziram through runoff and spray drift. PRZM simulates pesticide application, movement and transformation on an agricultural field and the resultant pesticide loadings to a receiving water body via runoff, erosion and spray drift. The EXAMS model simulates the fate of the pesticide and resulting concentrations in the water body. The standard scenario used for ecological pesticide assessments assumes application to a 10-hectare agricultural field that drains into an adjacent 1-hectare water body, 2-meters deep (20,000 m^3 volume) with no outlet. PRZM/EXAMS was used to estimate screening-level exposure of aquatic organisms to Ziram. The measure of exposure for aquatic species is the 1-in-10 year return peak or rolling mean concentration. The 1-in-10 year peak is used for estimating acute exposures of direct effects to the CRLF, as well as indirect effects to the CRLF through effects to potential prey items, including: algae, aquatic invertebrates, fish and frogs. The 1-in-10-year 60-day mean is used for assessing chronic exposure to the CRLF and fish and frogs serving as prey items; the 1-in-10-year 21-day mean is used for assessing chronic exposure for aquatic invertebrates, which are also potential prey items.

Exposure estimates for the terrestrial-phase CRLF and terrestrial invertebrates and mammals (serving as potential prey) assumed to be in the target area or in an area exposed to spray drift are derived using the T-REX model (version 1.3.1, 12/07/2006). This model incorporates the Kenega nomograph, as modified by Fletcher *et al.* (1994), which is based on a large set of actual field residue data. The upper limit values from the nomograph represented the 95th percentile of residue values from actual field measurements (Hoerger and Kenega, 1972). For modeling purposes, direct exposures of the CRLF to Ziram through contaminated food are estimated using the EECs for the small bird (20 g) consuming small insects. Dietary-based and dose-based exposures of potential prey (small mammals) are assessed using the small mammal (15 g) consuming short grass. The small bird (20g) consuming small insects and the small mammal (15g) consuming short grass are used because these categories represent the largest RQs of the size and dietary categories in T-REX that are appropriate surrogates for the CRLF and one of its prey items. Estimated exposures of terrestrial insects to Ziram are bound by using the dietary based EECs for small insects and large insects.

Birds are currently used as surrogates for terrestrial-phase CRLF. However, amphibians are poikilotherms (body temperature varies with environmental temperature) while birds are homeotherms (temperature is regulated, constant, and largely independent of environmental temperatures). Therefore, amphibians tend to have much lower metabolic rates and lower caloric intake requirements than birds or mammals. As a consequence, birds are likely to consume more food than amphibians on a daily dietary intake basis, assuming similar caloric content of the food items. Therefore, the use of avian food intake allometric equation as a surrogate to amphibians is likely to result in an over-estimation of exposure and risk for reptiles and terrestrial-phase amphibians. Therefore, T-REX (version 1.3.1) has been refined to the T-HERPS model (v. 1.0), which allows for an estimation of food intake for poikilotherms using the same basic procedure as T-REX to estimate avian food intake.

TerrPlant (version 1.2.2, 12/26/2006) is used to assess risk for indirect effects of critical habitat. EECs for terrestrial plants inhabiting dry and wetland areas are derived using seedling emergence and vegetative vigor toxicity endpoints for terrestrial plants. This model uses estimates of pesticides in runoff and in spray drift to calculate EECs. EECs are based upon solubility, application rate and minimum incorporation depth.

Spray drift models, AGDISP and/or AgDRIFT are used to assess exposures of terrestrial phase CRLF and its prey to ZIRAM deposited on terrestrial habitats by spray drift. AGDISP (version 8.13; dated 12/14/2004) (Teske and Curbishley, 2003) is used to simulate aerial and ground applications using the Gaussian farfield extension. In addition to the buffered area from the spray drift analysis, the downstream extent of Ziram that exceeds the LOC for the effects determination is also considered.

2.10.1.2 Measures of Effect

Data identified in Section 2.8 are used as measures of effect for direct and indirect effects to the CRLF. Data were obtained from registrant submitted studies or from literature studies identified by ECOTOX. The ECOTOXicology database (ECOTOX) was searched in order to provide more ecological effects data and in an attempt to bridge existing data gaps. ECOTOX is a source for locating single chemical toxicity data for aquatic life, terrestrial plants, and wildlife. ECOTOX was created and is maintained by the USEPA, Office of Research and Development, and the National Health and Environmental Effects Research Laboratory's Mid-Continent Ecology Division.

Although there are no toxicity values for amphibians from registrant submitted studies or open literature, representative species are used to estimate the effect of Ziram on the terrestrial and aquatic-phase CRLF. The assessment of risk for direct effects to the terrestrial-phase CRLF makes the assumption that toxicity of Ziram to birds is similar to or less than the toxicity to the terrestrial-phase CRLF. The same assumption is made for fish and aquatic-phase CRLF. Algae, aquatic invertebrates, fish, and amphibians represent potential prey of the CRLF in the aquatic habitat. Terrestrial invertebrates, small mammals, and terrestrial-phase amphibians represent potential prey of the CRLF in the terrestrial habitat. Aquatic, semi-aquatic, and terrestrial plants represent habitat of CRLF.

The acute measures of effect used for animals in this screening level assessment are the LD_{50}, LC_{50} and EC_{50}. LD stands for "Lethal Dose", and LD_{50} is the amount of a material, given all at once, that is estimated to cause the death of 50% of the test organisms. LC stands for “Lethal Concentration” and LC_{50} is the concentration of a chemical that is estimated to kill 50% of the test organisms. EC stands for “Effective Concentration” and the EC_{50} is the concentration of a chemical that is estimated to produce a specific effect in 50% of the test organisms. Endpoints for chronic measures of exposure for listed and non-listed animals are the NOAEL/NOAEC and NOEC. NOAEL stands for “No Observed-Adverse-Effect-Level” and refers to the highest tested dose of a substance that has been reported to have no harmful (adverse) effects on test organisms. The NOAEC (*i.e.*, “No-Observed-Adverse-Effect-Concentration”) is the highest test concentration at which none of the observed effects were statistically different from the control. The NOEC is the No-Observed-Effects-Concentration. For non-listed plants, only acute exposures are assessed (*i.e.*, EC_{25} for terrestrial plants and EC_{50} for aquatic plants).

It is important to note that the measures of effect for direct and indirect effects to the CRLF and its designated critical habitat are associated with impacts to survival, growth, and fecundity, and do not include the full suite of sublethal effects used to define the action area. According the Overview Document (USEPA 2004), the Agency relies on effects endpoints that are either direct measures of impairment of survival, growth, or fecundity or endpoints for which there is a scientifically robust, peer reviewed relationship that can quantify the impact of the measured effect endpoint on the assessment endpoints of survival, growth, and fecundity.

2.10.1.3 Integration of Exposure and Effects

Risk estimation (Section 5.1) is the integration of exposure and ecological effects to determine the potential ecological risk from agricultural and non-agricultural uses of Ziram, and the likelihood of direct and indirect effects to CRLF in aquatic and terrestrial habitats. The exposure and toxicity effects data are integrated in order to evaluate the risks of adverse ecological effects on non-target species. For the assessment of Ziram risks, the risk quotient (RQ) method is used to compare exposure and measured toxicity values. EECs are divided by acute and chronic toxicity values. The resulting RQs are then compared to the Agency's levels of concern (LOCs) (USEPA, 2004) (see E).

For this endangered species assessment, listed species LOCs are used for comparing RQ values for acute and chronic exposures of Ziram directly to the CRLF. If estimated exposures directly to the CRLF of Ziram resulting from a particular use are sufficient to exceed the listed species LOC, then the effects determination for that use is "may affect". When considering indirect effects to the CRLF due to effects to animal prey (aquatic and terrestrial invertebrates, fish, frogs, and mice), the listed species LOCs are also used. If estimated exposures to CRLF prey of Ziram resulting from a particular use are sufficient to exceed the listed species LOC, then the effects determination for that use is a "may affect." If the RQ being considered also exceeds the non-listed species acute risk LOC, then the effects determination is a LAA. If the acute RQ is between the listed species LOC and the non-listed acute risk species LOC, then further lines of evidence (*i.e*. probability of individual effects, species sensitivity distributions) are considered in distinguishing between a determination of NLAA and a LAA. When considering indirect effects to the CRLF due to effects to algae as dietary items or plants as habitat, the non-listed species LOC for plants is used because the CRLF does not have an obligate relationship with any particular aquatic and/or terrestrial plant. If the RQ being considered for a particular use exceeds the non-listed species LOC for plants, the effects determination is "may affect". Further information on LOCs is provided in Appendix E.

3. Exposure Assessment

Ziram is formulated as a dry flowable or water dispersable granular, both formulations are designed to be mixed with water and sprayed with ground or aerial equipment. Application equipment is not specified on the label, except as noted below. Risks from ground boom and aerial applications are considered in this assessment because they are expected to result in the highest off-target levels of Ziram due to generally higher spray drift levels. Ground boom and aerial modes of application tend to use lower volumes of application applied in finer sprays than applications coincident with sprayers and spreaders and thus have a higher potential for off-target movement via spray drift.

3.1 Label Application Rates and Intervals

Ziram labels may be categorized into two types: labels for manufacturing uses (including technical grade Ziram and its formulated products) and end-use products. While technical products, which contain Ziram of high purity, are not used directly in the environment, they are used to make formulated products, which can be applied in specific areas to control fungal diseases. The formulated product labels legally limit Ziram's potential use to only those sites that are specified on the labels.

Currently registered agricultural and non-agricultural uses of Ziram within California include almonds, apples/pears, apricots, cherries, grapes, peaches/nectarines, pecans and wine grapes. The uses assessed have been previously summarized in Table 7 and more specifics on how these were modeled are summarized in Table 15. Actual usage data for Ziram in California are summarized in Table 8.

3.2 Aquatic Exposure Assessment

3.2.1 Modeling Approach

Aquatic exposures are quantitatively estimated for all of assessed uses using scenarios that represent high exposure sites for Ziram use. Each of these sites represents a 10 hectare field that drains into a 1-hectare pond that is 2 meters deep and has no outlet. Exposure estimates generated using the standard pond are intended to represent a wide variety of vulnerable water bodies that occur at the top of watersheds including prairie pot holes, playa lakes, wetlands, vernal pools, man-made and natural ponds, and intermittent and first-order streams. As a group, there are factors that make these water bodies more or less vulnerable than the standard surrogate pond. Static water bodies that have larger ratios of drainage area to water body volume would be expected to have higher peak EECs than the standard pond. These water bodies will be either shallower or have large drainage areas (or both). Shallow water bodies tend to have limited additional storage capacity, and thus, tend to overflow and carry pesticide in the discharge whereas the standard pond has no discharge. As watershed size increases beyond 10 hectares, at some point, it becomes unlikely that the entire watershed is planted to a single crop, which is all treated with the pesticide. Headwater streams can also have peak concentrations higher than the standard pond, but they tend to persist for only short periods of time and are then carried downstream.

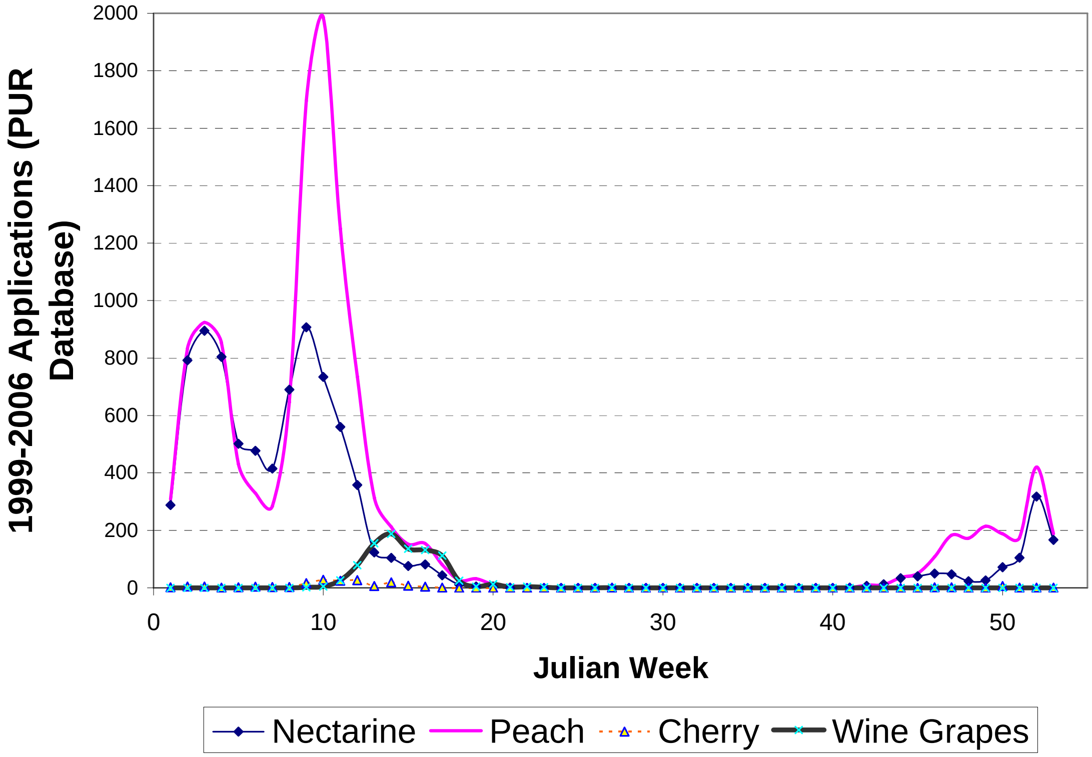


Figure 9. Number of applications of Ziram in California by week on four representative crops (Source: PUR database).

Crop-specific management practices for all of the assessed uses of Ziram were used for modeling, including application rates, number of applications per year, application intervals, buffer widths and resulting spray drift values modeled from AgDRIFT and AgDISP, and the first application date for each crop. The date of first application was developed based on several sources of information including data provided by BEAD, a summary of individual applications from the CDPR PUR data, and Crop Profiles maintained by the USDA. The timing of the applications was adjusted according to the predominant practice for the years 1999-2006 as included in the PUR Database (see Figure 9 for examples of the data used to make this determination). Detailed agronomic related inputs and the selected scenarios for aquatic exposure modeling are provided in Table 15.

Laboratory and field studies have shown Ziram is substantially converted to the more stable Thiram form in the environment. However, the conversion rate is irregular and, for chronic exposure, it was determined that PRZM/EXAMS modeling of combined residues does not provide higher estimates than a more conservative treatment involving modeling of chronic exposure to Thiram alone assuming 100% conversion of Ziram to Thiram on the day of each application. Therefore, the assessment of chronic exposure from Ziram uses is quantified as Thiram for chronic risk analyses.

3.2.2 Model Inputs

Ziram is a fungicide used on a variety of fruit and orchard crops in California (uses on ornamentals and a few other crops such as blackberries, blueberries and tomatoes are not registered in California). Environmental fate data are summarized and values for these parameters used in model input are identified in Table 13 for Ziram and in Table 14 for the degradate of toxicological concern, Thiram.

Table 13. Environmental fate data and selected values for PRZM/EXAMS model input for parent Ziram.

Parameter	Value	Calculation Source and Method*
Chemical name	Ziram	---
PC Code	034805	---
Molecular Weight	307.5	MRID 442284-01.
Solubility (mg/L or ppm)	65	Product chemistry data.
Henry's Law Constant	0	Agrochemical handbook, 3rd edition.
pH 5 Hydrolysis half life (days)	0.07 (NU)	MRID 43866701. Not used for model input.
pH 7 Hydrolysis half life (days)	0.74	MRID 43866701.
pH 9 Hydrolysis half life (days)	6.3 (NU)	MRID 43866701. Not used for model input.
Soil Photolysis half life (days)	0.3 (NU)	MRID 43642501.
Aquatic photolysis half life (days) (EXAMS KDP input)	0.43	MRID 44097701. Higher of 2 studies; both at pH 5.
Aerobic soil metabolism half life (days) (PRZM DWRATE input)	4.9	MRIDs 43985801; 46622302; 47005202. 90%ile upper C.I. on mean log linear first order $t_{1/2}$ from four studies.
Aerobic aquatic (water column) metabolism half life (days) (EXAMS KBACW input)	Stable	Guidance: If no aerobic aquatic metabolism data are available and there is significant hydrolysis, assume that the compound is stable to aquatic metabolism.
Anaerobic soil metabolism half life (days)	14.1	MRID 44228402. Not used in modeling.
Anaerobic aquatic (benthic) metabolism half life (days) (EXAMS KBACS input)	Stable	Guidance: If no anaerobic aquatic metabolism data are available and significant hydrolysis occurs, assume that the compound is stable to aquatic metabolism.

Parameter	Value	Calculation Source and Method*
K_d or K_f (ml/g) (PRZM KD input)	5.7 (input value) (Range 2.9-68.1)	MRID 43873501. Lowest non- sand K_f (K_{oc} highly variable between 4 test soils).
K_{oc} (ml/g)	314-3732	MRID 43873501. Not used for model input.
Application Rates	See Table 15.	Product labels
Application Efficiency	0.95 (aerial) 0.99 (ground)	Per Guidance
Spray drift mass input to pond	0.05 (aerial) 0.01 (ground)	Per Guidance
Number of Applications	See Table 15.	Product label.
Application Method	Foliar spray; ground or aerial	Product label.
Flag for Runoff flow	None	Per guidance in PE-5 User's Manual. http://www.epa.gov/oppefed1/models/water/pe5_user_manual.htm
Chemical Application Method (PRZM CAM input)	2	CAM 2 = interception based on crop canopy, as a straight-line function of crop development; chemical reaching the soil surface is incorporated to 4 cm;
Incorporation Depth (cm) – Incorporation Depth (cm) –	0	NA
IPSCND (post-harvest foliar pesticide disposition)	1	Flag indicating the disposition of pesticide remaining on foliage after harvest. This flag only applies if CAM = 2 or 3. If IPSCND = 1, pesticide remaining on foliage is converted to surface application to the top soil layer. If IPSCND = 2, remaining pesticide on foliage is completely removed after harvest. If IPSCND = 3, remaining pesticide on foliage is retained as surface residue and continues to undergo decay.
Field Size	EPA Pond	Uses a 10-hectare field draining into the pond.

* "Guidance" refers to the "Guidance for Selecting Input Parameters in Modeling the Environmental Fate and Transport of Pesticides": Version II dated February 28, 2002; unless otherwise specified. See: http://www.epa.gov/oppefed1/models/water/input_guidance2_28_02.htm

Table 14. Environmental fate data and selected values for PRZM/EXAMS model input for Thiram (as a Ziram metabolite).

Chemical name	Thiram	Calculation Source and Method*
PC Code	079801	---
Molecular Weight	240.44	Product chemistry data
Solubility (mg/L or ppm)	16	Product chemistry data
Henry's Law Constant	0	Agrochemical handbook, 3rd edition.
pH 5 Hydrolysis half life (days)	68.50; 169.00	MRIDs 45714101 and 41840601. Not used for model input.
pH 7 Hydrolysis half life (days)	17.9 (measured value of 3.5 days not used for model input)	MRIDs 45714101 and 41840601. Most conservative of values measured in two studies (both classified as supplemental); $t_{1/2}$ =3.5 days in other study.
pH 9 Hydrolysis half life (days)	0.29; 6.90	MRIDs 45714101 and 41840601. Not used for model input
Soil Photolysis half life (days)	0.72	MRID 45724501. Not used for model input.
Aquatic photolysis half life (days) (EXAMS KDP input)	0.36	MRID 41753801.
Aerobic soil metabolism half life (days) (PRZM DWRATE input)	17.2	Over all log linear first order $t_{1/2}$ (Not multiplied by 3 because inference from multiple Ziram studies is that the $t_{1/2}$ is typically shorter than 17 days.
Aerobic aquatic (water column) metabolism half life (days) (EXAMS KBACW input)	2.5	MRID 45243401. 90%ile upper C.I. on mean log linear first order $t_{1/2}$ from two studies.
Anaerobic soil metabolism half life (days)	No valid study.	Not used in modeling.
Anaerobic aquatic (benthic) metabolism half life (days) (EXAMS KBACS input)	129.3	MRID 43628501. Guidance: If only one half-life value is available, use 3x the measured $t_{1/2}$ (43.1 days in this case).
K_d or K_f (ml/g) (PRZM KD input)	54 (model input) (54 to 267 in 4 test soils)	MRID 43787501. Lowest non- sand K_f (K_{oc} highly variable between 4 test soils.)
K_{oc} (ml/g)	2245 to 24526	MRID 43787501. Not used for model input.
Application Rates	See Table 15..	Product labels.
Application Efficiency	0.95 (aerial) 0.99 (ground)	Per Guidance

Chemical name	Thiram	Calculation Source and Method*
Spray drift mass input to pond	0.05 (aerial) 0.01 (ground)	Per Guidance
Number of Applications	See Table 15..	Product label.
Application Method	Foliar spray; ground or aerial	Product label.
Flag for Runoff flow	None	Per guidance in PE-5 User's Manual. http://www.epa.gov/oppefed1/models/water/pe5_user_manual.htm
Chemical Application Method (PRZM CAM input)	2	CAM 2 = interception based on crop canopy, as a straight-line function of crop development; chemical reaching the soil surface is incorporated to 4 cm;
IPSCND (post-harvest foliar pesticide disposition)	1	Flag indicating the disposition of pesticide remaining on foliage after harvest. This flag only applies if CAM = 2 or 3. If IPSCND = 1, pesticide remaining on foliage is converted to surface application to the top soil layer. If IPSCND = 2, remaining pesticide on foliage is completely removed after harvest. If IPSCND = 3, remaining pesticide on foliage is retained as surface residue and continues to undergo decay.
Field Size	EPA Pond	Uses a 10-hectare field draining into the pond.

* "Guidance" refers to the "Guidance for Selecting Input Parameters in Modeling the Environmental Fate and Transport of Pesticides": Version II dated February 28, 2002; unless otherwise specified. See: http://www.epa.gov/oppefed1/models/water/input_guidance2_28_02.htm

Table 15. Agronomic inputs used in aquatic exposure assessments with PRZM-EXAMS.

Use Site[1]	Single applic. Rate (lb ai/A)	# Applic.	Rate / Year (lb ai/A)	First Applic. Date[4]	Interval	Last Date	Selected Scenario / Comments
Almonds	6.1	4	24.4	03 Mar	7	24 Mar	CA almond STD
Apples/Pears (West)	4.6	4	18.4	17 Mar (pears) 7 Apr (apples)	7	07-Apr	CA fruit STD Aerial application allowed only as pre-harvest application
Apricots	6.1	4	24.4	18 Feb	7	10 Mar	CA fruit STD
Cherries (CA)	3.8	4	15.2	3 Mar	7	24 Mar	CA fruit STD
Grapes (West)[2]	3.0	6 (assumed)	18 (assumed)	17 Mar	7	21 Apr	CA fruit STD (same runs as pears / apples)
Grapes –wine (West)	3.0	6 (assumed)	18 (assumed)	24 Mar	7	28 Apr	CA wine grapes RLF
Nectarines (Western US for leaf curl)	7.6	6	45.6	07 Jan	7	11 Feb	CA fruit STD
Peaches (Western US for leaf curl)	7.6	6	45.6	25 Feb	7	31 Mar	CA fruit STD
Pecans[3] (no use reported in CA)	6.1	6	36.6	15 Feb	7	21 Mar	CA almond STD Aerial application is prohibited.

[1] CA = Agronomic model inputs based on label directions for California only.

[2] West = Agronomic model inputs based on label directions for crops grown to the west of the Rockies. No specification is for crops grown anywhere in the US.

[3] Although this use is registered in California, no use of Ziram was reported on pecans in California for the period 1999-2006.

[4] The "multi-run" feature of PE-5 was used to batch process PRZM-EXAMS with three sets of application dates. The "first application date" represents the initial application date used in the first PRZM-EXAMS run, this was followed by two more model runs with the initial application date incrementally moved to three days later; application intervals of 7 days were always used except as noted. For example, the following date sets were used for almonds:
Model run #1: Mar 3, Mar 10, Mar 17, Mar 24.
Model run #2: Mar 6, Mar 13, Mar 20, Mar 27.
Model run #3: Mar 9, Mar 16, Mar 23, Mar 30.

3.2.3 Results

The aquatic EECs for the various scenarios and application practices are listed in Table 16. In general, exposure tends to be higher for the crops where applications typically begin in the winter. All scenarios were run with 3 different sets of application dates and the average of the 1 in 10 year return frequency concentrations from the three runs was averaged. Because of this and because the timing fit closely with reported use history data from the PUR database, the results are believe to well represent what might occur with each crop use. The chronic EECs are for Thiram formed from Ziram are conceptionally higher than they would be for Ziram because of its shorter persistence in the environment. Therefore Thiram EECs are used in the assessment to provide the more conservative estimate of risk. In the context of applications of Ziram, the Thiram estimates are conservative (i.e., should be higher than actually occur) in that the modeling assumes 100% instantaneous conversion from Ziram to Thiram. Detailed results from PRZM/EXAMS modeling are provided in Appendix F.

Table 16. Aquatic EECs (μg/L) for ZIRAM Uses in California.

Compound used as basis for modeling	Equipment	Crop	peak	21day	60day
Ziram	Ground	Almonds	16.75	NU	NU
Ziram	Aerial	Almonds	23.44	NU	NU
Ziram	Aerial	Apples	15.14*	NU	NU
Ziram	Ground	Apricots	23.51	NU	NU
Ziram	Aerial	Apricots	32.94	NU	NU
Ziram	Ground	Cherries	9.89	NU	NU
Ziram	Aerial	Cherries	13.31	NU	NU
Ziram	Ground	Grapes	13.47	NU	NU
Ziram	Aerial	Grapes	15.82	NU	NU
Ziram	Ground	Nectarines	34.61	NU	NU
Ziram	Aerial	Nectarines	42.16	NU	NU
Ziram	Ground	Peaches	32.58	NU	NU
Ziram	Aerial	Peaches	35.64	NU	NU
Ziram	Ground	Pears	14.94	NU	NU
Ziram	Ground	Pecans	28.63	NU	NU
Ziram	Ground	Wine Grapes	43.70	NU	NU
Ziram	Aerial	Wine Grapes	42.46	NU	NU
Thiram	Aerial	Almonds	NU	14.47	7.53
Thiram	Aerial	Apples	NU	8.49	3.37
Thiram	Aerial	Apricots	NU	14.06	7.22
Thiram	Aerial	Cherries	NU	8.62	4.29
Thiram	Aerial	Grapes	NU	6.67	5.36
Thiram	Aerial	Nectarines	NU	24.38	16.35
Thiram	Aerial	Peaches	NU	16.13	10.86
Thiram	Aerial	Pears	NU	2.32	1.25
Thiram	Ground	Pecans	NU	5.72	3.59

Compound used as basis for modeling	Equipment	Crop	peak	21day	60day
Thiram	Aerial	Wine Grapes	NU	13.69	8.53

Notes:
Generally, Thiram EECs were only calculated for the aerial applications as the parent Ziram EECs were higher for aerial (when permitted) than ground applications.
The usage directions on labels are exactly the same for apples and pears; simulations are designated as representing "apples" or "pears" depending on which of these crops the chosen application dates for the model simulations most closely represented.
* The simulation for aerial application to apples does not correspond exactly to the labeled use; only a pre-harvest application may be made aerially; this type of applications appears also to be uncommon in California.

3.2.4 Existing Monitoring Data

A critical step in the process of characterizing EECs is comparing the modeled estimates with available surface water monitoring data. However, no monitoring data are available for either Ziram or its degradate Thiram from the USGS NAWQA program (*http://water.usgs.gov/nawqa*). Also, no monitoring data are available from the California Department of Pesticide Regulation databases (see http://www.cdpr.ca.gov/docs/emon). No air monitoring data were located.

3.2.5 Spray Drift Buffer Analysis

The action area is the entire state of California (due to the carcinogenic endpoint). An analysis of spray drift buffers can be found in the Spatial Analysis Section 5.3.

3.2.6 Downstream Dilution Analysis

Although the initial action area is the entire state of California, an analysis of the maximum amount of downstream dilution was performed based off-site transport. The spatial extent of the effects determination is based on the initial area of concern for application of Ziram on orchards/vineyards and expanded to include the total area where there is potential for direct or indirect effects to occur via off-site transport mechanisms. The downstream extent analysis shows that 225 kilometers is the furthest distance that could be added downstream. This distance is representative of the maximum continuous downstream dilution from the edge of the initial area of concern where direct/indirect effects and/or critical habitat modification may occur. Lotic (*i.e.*, flowing) waterbodies that overlap with the CRLF habitat potentially contain concentrations of Ziram sufficient to result in LAA determination and modification of critical habitat.

3.3 Terrestrial Animal Exposure Assessment

T-REX (Version 1.3.1) is used to calculate dietary and dose-based EECs of Ziram for the CRLF and its potential prey (*e.g.* small mammals and terrestrial insects) inhabiting terrestrial areas. EECs used to represent the CRLF are also used to represent exposure values for frogs serving as potential prey of CRLF adults. T-REX simulates a 1-year time period. For this assessment, spray applications of water dispersable formulations of Ziram are considered, as discussed in below.

Terrestrial EECs for foliar applications of Ziram were derived for the uses summarized in Table 17. Given that no data on interception and subsequent dissipation from foliar surfaces is available for Ziram, a default foliar dissipation half-life of 35 days is used based on the work of Willis and McDowell (1987) Use specific input values, including number of applications, application rate and application interval are provided in Table 17. An example output from T-REX is available in Appendix **G**.

Table 17. Input Parameters for Foliar Applications Used to Derive Terrestrial EECs for Ziram with T-REX.

Use (Application method)	Application rate (lbs ai/A)	Number of Applications
Almonds	6.1	4
Apples/pears	4.6	4
Apricots	6.1	4
Cherries	3.8	4
Grapes/Wine grapes	3	7
Peaches/nectarines	7.6	6
Pecans	6.1	6

T-REX is also used to calculate EECs for terrestrial insects exposed to Ziram. Dietary-based EECs calculated by T-REX for small and large insects (units of a.i./g) are used to calculate the bounds for exposure to bees. Available acute contact toxicity data for bees exposed to Ziram (in units of µg a.i./bee) are converted to µg a.i./g (of bee) by multiplying by 1 bee/0.128 g. The EECs are later compared to the adjusted acute contact toxicity data for bees in order to derive RQs.

Table 18 presents the EECs for both Ziram and Thiram. EECs for small insects range from 1464 ppm for cherries to 5213 ppm for nectarines/peaches. EECs for large insects range from 163 ppm for cherries to 579 ppm for nectarines/peaches.

Table 18. EECs (ppm) for Indirect Effects of Ziram and Thiram to the Terrestrial-Phase CRLF via Effects to Terrestrial Invertebrate Prey Items

Use	Ziram		Thiram	
	Small Insect	Large Insect	Small Insect	Large Insect
Almonds	3391	377	3020	335
Apples/Pears	2245	249	2277	253
Apricots	3391	377	3020	335

Cherries	1464	163	1881	209
Grapes	2058	229	1943	216
Nectarines/Peaches	5213	579	5334	593
Pecans	4184	465	2221	247

For modeling purposes, exposures of the CRLF to Ziram through contaminated food are estimated using the EECs for the small bird (20 g) consuming small insects. Dietary-based and dose-based exposures of potential prey are assessed using the small mammal (15 g) consuming short grass. Upper-bound Kenega monogram values reported by T-REX for these two organism types are used for derivation of EECs for the CRLF and its potential prey. Table 18 presents EECs for terrestrial invertebrates small and large insects reported by T-REX. The resulting adjusted EECs for birds and small mammals are available in Table 19. The diet of the CRLF also includes fish as prey (See Table 16 for aquatic EECs for fish as prey). An example output from T-REX v. 1.3.1 is available in **G**.

Table 19. Upper-bound Kenega Nomogram EECs for Dietary- and Dose-based Exposures of the CRLF and its Prey to Thiram.

Use	EECs for CRLF		EECs for Prey (small mammals)	
	Dietary-based EEC (ppm)	Dose-based EEC (mg/kg-bw)	Dietary-based EEC (ppm)	Dose-based EEC (mg/kg-bw)
Almonds	2977.64	3391.23	2977.64	5047.03
Apples/Pears	2245.43	2557.32	2245.43	3805.95
Apricots	2977.64	3391.23	2977.649	5047.03
Cherries	1464.41	1667.82	1464.41	2482.14
Grapes	2057.72	2343.54	2057.72	3487.79
Peaches	5212.90	5936.97	5212.90	4970.10
Pecans	4184.04	4765.20	4184.04	7091.84

3.4 Terrestrial Plant Exposure Assessment

Although the maximum label rate is 7.6 lb/A for nectarines/peaches, the terrestrial plant studies were conducted with one concentration of 6.1 lb/A, near the label rate. No EC50 was estimated from the toxicity studies due to no treatment-related effects at the highest tested concentration observed for either the seedling emergence or vegetative vigor studies. Therefore, no effects to terrestrial plants are expected. . Based on the results of the seedling emergence and vegetative studies, no RQs will be estimated using the TerrPlant model.

4. Effects Assessment

This assessment evaluates the potential for ZIRAM to directly or indirectly affect the CRLF or modify its designated critical habitat. As previously discussed in Section 2.7, assessment endpoints for the CRLF effects determination include direct toxic effects on the survival, reproduction, and growth of CRLF, as well as indirect effects, such as reduction of the prey base or modification of its habitat. In addition, potential modification of critical habitat is assessed by evaluating effects to the PCEs, which are components of the critical habitat areas that provide essential life cycle needs of the CRLF. Direct effects to the aquatic-phase of the CRLF are based on toxicity information for freshwater fish, while terrestrial-phase effects are based on avian toxicity data, given that birds are generally used as a surrogate for terrestrial-phase amphibians. Because the frog's prey items and habitat requirements are dependent on the availability of freshwater fish and invertebrates, small mammals, terrestrial invertebrates, and aquatic and terrestrial plants, toxicity information for these taxa are also discussed. Acute (short-term) and chronic (long-term) toxicity information is characterized based on registrant-submitted studies and a comprehensive review of the open literature on Ziram.

As described in the Agency's Overview Document (U.S. EPA, 2004), the most sensitive endpoint for each taxa is used for risk estimation. For this assessment, evaluated taxa include aquatic-phase amphibians, freshwater fish, freshwater invertebrates, aquatic plants, birds (surrogate for terrestrial-phase amphibians), mammals, terrestrial invertebrates, and terrestrial plants.

Toxicity endpoints are established based on data generated from guideline studies submitted by the registrant, and from open literature studies that meet the criteria for inclusion into the ECOTOX database maintained by EPA/Office of Research and Development (ORD) (U.S. EPA, 2004). Open literature data presented in this assessment were obtained from ECOTOX information obtained on [February 28, 2008]. In order to be included in the ECOTOX database, papers must meet the following minimum criteria:

(1) the toxic effects are related to single chemical exposure;
(2) the toxic effects are on an aquatic or terrestrial plant or animal species;
(3) there is a biological effect on live, whole organisms;
(4) a concurrent environmental chemical concentration/dose or application rate is reported; and
(5) there is an explicit duration of exposure.

Data that pass the ECOTOX screen are evaluated along with the registrant-submitted data, and may be incorporated qualitatively or quantitatively into this endangered species assessment. In general, effects data in the open literature which are more conservative than the registrant-submitted data are considered. The degree to which open literature data are quantitatively or qualitatively characterized for the effects determination is dependent on whether the information is relevant to the assessment endpoints (*i.e.*, maintenance of CRLF survival, reproduction, and growth) identified in Section 2.8. For example, endpoints such as behavior modifications are likely to be qualitatively evaluated, because quantitative relationships between modifications and reduction in species survival, reproduction, and/or growth are not available. Although the effects determination relies on endpoints that are relevant to the assessment endpoints of survival, growth, or reproduction, it is important to note that the full suite of sub-lethal endpoints potentially available in the effects literature (regardless of their significance to the assessment endpoints) are considered to define the action area for Ziram.

Citations of all open literature not considered as part of this assessment because they were either rejected by the ECOTOX screen or accepted by ECOTOX but not used (e.g., the endpoint is less sensitive) are included in **Appendix H.** **Appendix H** also includes a rationale for rejection of those studies that did not pass the ECOTOX screen and those that were not evaluated as part of this endangered species risk assessment.

A detailed spreadsheet of the available ECOTOX open literature data, including the full suite of lethal and sub-lethal endpoints is presented in Appendix **I**. Appendix J also includes a summary of the human health effects data for Ziram.

In addition to registrant-submitted and open literature toxicity information, other sources of information, including use of the acute probit dose response relationship to establish the probability of an individual effect and reviews of the Ecological Incident Information System (EIIS), are conducted to further refine the characterization of potential ecological effects associated with exposure to Ziram. A summary of the available aquatic and terrestrial ecotoxicity information, use of the probit dose response relationship, and the incident information for Ziram are provided in Sections 4.1 through 4.4, respectively.
A detailed summary of the available ecotoxicity studies for Ziram is presented in **C.**

Ziram degrades into Thiram, which is more persistent. Both are present over short periods, however, Ziram is more toxic based on acute aquatic toxicity studies. Therefore acute toxicity studies for Thiram are presented in D.

Due to Thiram's persistence, chronic toxicity values will be based on Thiram modeling to calculate RQs for chronic risk. The methodology used, which was based upon 100% conversion of the applied Ziram to Thiram insured that the exposure estimates were conservative (protective) for any scenario of chronic exposure to residues from Ziram applications.

In addition to reviewing registrant submitted studies, open literature, incident reports from the EIIS database, as well as any information about mixtures is reviewed. Open literature and incident reports are identified in Section 4 for each species. There is one registered mixture product reported for Ziram (Polyphase 685).

4.1 Toxicity of Ziram to Aquatic Organisms

Table 20 summarizes the most sensitive aquatic toxicity endpoints for the CRLF, based on an evaluation of both the submitted studies and the open literature, as previously discussed for Ziram . Table 23 summarizes the most sensitive endpoints for the CRLF for the degradate Thiram. A brief summary of submitted and open literature data considered relevant to this ecological risk assessment for the CRLF is presented below. Additional information is provided in Appendix C and Appendix D**.**

Table 20. Freshwater Aquatic Toxicity Profile.

Assessment Endpoint	**Ziram** Species	Toxicity Value Used in Risk Assessment	Citation MRID # (Author & Date)	**Thiram** Species	Toxicity Value Used in Risk Assessment	Citation MRID # (Author & Date)
Acute Direct Toxicity to Aquatic-Phase CRLF	(*Lepomis macrochirus*) Bluegill	LC50=9.7µg /L Acceptable	423863-03	(*Rasbar heteromorpha*) Harlequinn (TEP)	LC50= 7µg/L Supplemental	00034713
				(*Lepomis macrochirus*) Bluegill	LC50=42 µg a.i./L Acceptable	070801
	(Oncorhynchus mykiss) Trout	LC50= 1700 µg/L Acceptable	423863-04			
	Pimephales promelas Fathead Minnow	LC50= 8 µg/L Supplemental	Maloney and Palmer (1956) Water and Sewage Works 103:509-513 (ID 5003523)			
Chronic Direct Toxicity to Aquatic-Phase CRLF	*Pimephales promelas* Fathead minnow	NOAEC= 101 µg/L Most sensitive endpoint was post-hatch survival.		*Pimephales promelas*	ACR=530.2 µg a.i./L	
Indirect Toxicity to Aquatic-Phase CRLF via Acute Toxicity to Freshwater Invertebrates (i.e. prey items)	*Daphnia magna*	LC50= 48 µg/L Acceptable	423863-05	*Daphnia*	210 *Psuedokirchneriella subcapitat*	164662
Indirect Toxicity to Aquatic-	*Daphnia magna*	21 day NOAEC 39	MRID 468233-01	*Daphnia*	ACR=170.6	

	Ziram			Thiram		
Assessment Endpoint	**Species**	**Toxicity Value Used in Risk Assessment**	**Citation MRID # (Author & Date)**	Species	**Toxicity Value Used in Risk Assessment**	**Citation MRID # (Author & Date)**
Phase CRLF via Chronic Toxicity to Freshwater Invertebrates (i.e. prey items)		= µg/L 21 day LOAEC = 77 µg/L Most sensitive endpoint was length.			µg a.i./L	
Indirect Toxicity to Aquatic-Phase CRLF via Acute Toxicity to Non-vascular Aquatic Plants	*Psuedokirchneriella subcapitat (formerly) Selenestrum capricornum*	EC50=0.067 mg/L	MRID 438339-01	*Psuedokirchneriella subcapitat*	140 µg a.i./L	440861-01
Indirect Toxicity to Aquatic-Phase CRLF via Acute Toxicity to Vascular Aquatic Plants	*Lemna gibba G3*	Biomass EC50=0.37 mg/L (0.27-0.51mg/L0 Number of fronds EC50 = 0.71mg/L (0.45-1.1mg/L) Frond Count and Biomass NOAEC = 0.0351mg/L Biomass EC500.37mg/L (0.27-0.51mg/L) NOAEC=0.0351mg/L Acceptable	MRID 468233-02	*Lemna gibba G3*	EC50=1600 *Lemna gibba G3* Number of fronds EC50 = 1.6mg/L (1.3-1.9mg/L)	454412-02

[1] Including study acceptability and/or level of effect (LOAEC and endpoints on which the LOAEC was based for chronic effects.

Toxicity to aquatic fish and invertebrates is categorized using the system shown in Table 21 (U.S. EPA, 2004). Toxicity categories for aquatic plants have not been defined.

Table 21. Categories of Acute Toxicity for Aquatic Organisms.

LC_{50} (ppm)	Toxicity Category
< 0.1	Very highly toxic
> 0.1 - 1	Highly toxic
> 1 - 10	Moderately toxic
> 10 - 100	Slightly toxic
> 100	Practically nontoxic

4.1.1 Toxicity to Freshwater Fish

Given that no Ziram toxicity data are available for aquatic-phase amphibians, freshwater fish data were used as a surrogate to estimate direct acute and chronic risks to the CRLF. Freshwater fish toxicity data were also used to assess potential indirect effects of Ziram to the CRLF. Effects to freshwater fish resulting from exposure to Ziram have the potential to indirectly affect the CRLF via reduction in available food. As discussed in Section 2.5.3, over 50% of the prey mass of the CRLF may consist of vertebrates such as mice, frogs, and fish (Hayes and Tennant, 1985).

A summary of acute and chronic freshwater fish data, including data from the open literature, is provided below in Sections 4.1.1.1 through 4.1.1.3. The toxicity endpoints used to calculate RQs are identified in each section.

4.1.1.1 Freshwater Fish: Acute Exposure (Mortality) Studies

The toxicity values classify Ziram as moderately (MRID 423863-04) to very highly toxic (MRID 423863-03) to freshwater fish on an acute exposure basis.. The most sensitive acceptable acute freshwater fish study submitted (MRID 423863-03) resulted in an LC50=0.0097 mg/L.

4.1.1.2 Freshwater Fish: Chronic Exposure (Growth/Reproduction) Studies

Due to the absence of chronic data for Thiram, the acute-to-chronic (ACR) method will be used to estimate a chronic value. The chronic Thiram NOAEC is based on [the Ziram chronic fish NOAEC=101 µg/L divided by the fish LC50=8 µg/L] multiplied by the Thiram acute fish LC50=42 µg/L. Therefore, the Thiram chronic fish NOAEC is 530.25 µg/l. This calculation using this value will be described in the risk estimation section of the assessment.

4.1.1.3 Freshwater Fish Open Literature

However, the results from an ecotox study classified as supplemental (ID5003523) resulted in an LC_{50}=0.008 mg/L for the fathead minnow. The most sensitive toxicity value for the fathead minnow (LC50=8µg/L) will be used in the assessment.

4.1.2 Toxicity to Freshwater Invertebrates

Freshwater aquatic invertebrate toxicity data were used to assess potential indirect effects of Ziram to the CRLF. Effects to freshwater invertebrates resulting from exposure to Ziram have the potential to indirectly affect the CRLF via reduction in available food items. As discussed in Section 2.5.3, the main food source for juvenile aquatic- and terrestrial-phase CRLFs is thought to be aquatic invertebrates found along the shoreline and on the water surface, including aquatic sowbugs, larval alderflies and water striders.

4.1.2.1 Freshwater Invertebrates: Acute Exposure Studies

The 48-hour acute toxicity test (MRID 423863-05) using *Daphnia magna* resulted in an LC_{50}=48 µg/l. This classifies Ziram as very highly toxic to aquatic invertebrates. This toxicity value will be described in the risk estimation section.

In addition to the acute Ziram toxicity values, the acute toxicity values for Thiram will be used as additional evidence for the risk determination. The Thiram acute toxicity studies using *Daphnia* EC50=210 µg/L classifies Thiram as highly toxic to aquatic invertebrates. The acute Thiram EC50=210 µg/L will also be used in the acute-to-chronic ratio method to estimate the chronic value to be described in the risk estimation section.

4.1.2.2 Freshwater Invertebrates: Chronic Exposure Studies

Due to the absence of chronic data for Thiram, the acute-to-chronic (ACR) method will be used to estimate a chronic value for aquatic invertebrates. The chronic Thiram NOAEC is based on [the Ziram chronic *Daphnia* NOAEC=39 µg/L divided by the *Daphnia* LC50=48 µg/L] multiplied by the Thiram acute *Daphnia* EC50=210 µg/L. Therefore, the Thiram chronic *Daphnia* NOAEC is 170.6 µg/l. This calculation using this value will be described in the risk estimation section of the assessment. Studies used to estimate the chronic Thiram value are described in Appendix D.

4.1.2.3 Freshwater Invertebrates: Open Literature Data

In addition to reviewing registrant submitted toxicity studies, open literature is also reviewed for more sensitive endpoints to be included in the assessment. There was no open literature identified with more sensitive toxicity values available for Ziram.

4.1.3 Toxicity to Aquatic Plants

Aquatic plant toxicity studies were used as one of the measures of effect to evaluate whether Ziram may affect primary production and the availability of aquatic plants as food for CRLF tadpoles. Primary productivity is essential for indirectly supporting the growth and abundance of the CRLF.

Two types of studies were used to evaluate the potential of Ziram to affect aquatic plants. Laboratory and field studies were used to determine whether Ziram may cause direct effects to aquatic plants. A summary of the laboratory data and freshwater field studies for aquatic plants is provided in Sections 4.1.3.1 and 4.1.4.

4.1.3.1 Aquatic Plants: Vascular Plants

To estimate the effect of Ziram on the aquatic habitat for the CRLF, aquatic vascular studies were reviewed. In a 7-day acute toxicity study, the freshwater floating aquatic vascular plants Duckweed (*Lemna gibba* G3) were exposed to Ziram Technical at nominal concentrations of 0 (negative and solvent controls), 0.019, 0.041, 0.090, 0.20, 0.44, 0.96, 2.1 and 4.6 mg/L under static renewal conditions. The NOAEC and EC_{50} values based on biomass, the most sensitive endpoint, were 0.0351 and 0.37 mg/L, respectively. The percent growth inhibition, based on frond number, in the treated culture as compared to the control ranged from -1.7 to 72.5%. The percent growth inhibition, based on biomass, in the treated culture as compared to the control ranged from -7.1 to 90.6%. Observed effects included breaking up of colonies, root destruction, small fronds, chlorosis and necrosis. Effects were isolated to the measured 0.178-4.52 mg/L treatment groups. The results from this toxicity study will be described in the risk estimation section of the assessment.

A summary of the Thiram toxicity study using *Lemna* is found in Appendix D. The Thiram toxicity value (1.6 mg/L for frond number) from the registrant submitted study for the degradate is less sensitive than the parent

4.1.3.2 Aquatic Plants: Nonvascular Plants

The toxicity value for Ziram for the non-vascular aquatic plant *Pseudokirchneriella subcapitata* (formerly *Selenastrum capricornutum*) was EC50=67 µg/L (MRID438339-01). The toxicity value for Ziram will be described in the risk estimation section.

In addition to the non-vascular toxicity plant studies for the parent Ziram, there were also toxicity studies for Thiram. The toxicity value for Thiram for the non-vascular aquatic plant *Pseudokirchneriella subcapitata* (formerly *Selenastrum capricornutum)* was EC50=140 µg/L (MRID 440861-01. The RQs for Thiram are described in the risk description section as additional lines of evidence.

4.1.3.3 Open Literature for Aquatic Plants

In addition to reviewing registrant submitted aquatic plant studies, a search for open literature in ECOTOX was performed. No open literature with more sensitive endpoints for aquatic plants was available for Ziram.

4.1.4 Freshwater Field/Mesocosm Studies

Registrant submitted studies and open literature were reviewed to determine if there were any freshwater field or mesocosm studies for Ziram or Thiram. No freshwater field or mesocosm studies are available.

4.2 Toxicity of Ziram to Terrestrial Organisms

Table 22 summarizes the most sensitive terrestrial toxicity endpoints for the CRLF, based on an evaluation of both the submitted studies and the open literature. A brief summary of submitted and open literature data considered relevant to this ecological risk assessment for the CRLF is presented below. The most sensitive endpoint for either Ziram or Thiram is highlighted in bold

Table 22. Terrestrial Toxicity Profile.

	Ziram			**Thiram**		
Endpoint	**Species**	**Toxicity Value Used in Risk Assessment**	**Citation MRID# (Author & Date)**	**Species**	**Toxicity Value Used in Risk Assessment**	**Citation**
Acute Direct Toxicity to Terrestrial-Phase CRLF (LD_{50})	(*Colinua virginianus*) Quail	LD50=97 ppm	417257-01	(*Anas platyrhynchus*) Mallard	>2800mg/kg	BAOTHI03
Acute Direct Toxicity to Terrestrial-Phase CRLF (LC_{50})	(*Anas platyrhynchus*) Mallard	LC50=5156 ppm	423863-02	(*Anas platyrhynchus*) Mallard	>5,000 mg/kg	022923
				(*Colinua virginianus*) Quail	LC=3950 mg/kg	022923
Chronic Direct Toxicity to Terrestrial-Phase CRLF	(Anas platyrhynchus) Mallard	NOAEL=29 mg/kg	472865-01	(*Colinua virginianus)* Quail	NOAEC=9.6mg/kg/day (50% reduction in egg production)	454412-01 Acceptable
Indirect Toxicity to Terrestrial-Phase CRLF (via acute toxicity to mammalian prey items)	(Rattus norvegicus) Rat	LD50=320 ppm	413404-01	(Rattus norvegicus) Rat	LD50=2600 mg/kg	00153548

	Ziram			Thiram		
Endpoint	**Species**	**Toxicity Value Used in Risk Assessment**	**Citation MRID# (Author & Date)**	**Species**	**Toxicity Value Used in Risk Assessment**	**Citation**
Indirect Toxicity to Terrestrial-Phase CRLF (via chronic toxicity to mammalian prey items)	(Rattus norvegicus) Rat	**NOAEC=207 ppm**	439358-1			
Indirect Toxicity to Terrestrial-Phase CRLF (via acute toxicity to terrestrial invertebrate prey items)	(Apis mellifera) Honey bee	LD50>100 µg/bee	**416679-01**	(Apis mellifera) Honey Bee	LD50= 73.72	0003635
↑ Indirect Toxicity to Terrestrial- and Aquatic-Phase CRLF (via toxicity to terrestrial plants) ↓	Seedling Emergence Monocots	EC25 >6lbs/A NOAEC=6lbs/A. Purity=76.6%. No treatment effects were observed for any concentration.	MRID 468931-01 Acceptable.	NO DATA	NO DATA	NO DATA
	Seedling Emergence Dicots	EC25 >6lbs/A NOAEC=6lbs/A Purity=76.6%. Most sensitive dicot was soybean based on 16% reduction relative to negative control.	MRID 468931-01 Acceptable.	NO DATA	NO DATA	NO DATA
	Vegetative Vigor Monocots	EC25>6 lbs/A NOAEC<6 lbs/A Most sensitive monocot was rye grass with a 13% reduction in comparison to the control.	MRID 468931-01 Acceptable	NO DATA	NO DATA	NO DATA
	Vegetative Vigor Dicots	EC25>6 lbs/A NOAEC=6 lbs/A Most sensitive dicot was tomato with a 12% reduction in comparison to the control.	MRID 468931-01 Acceptable	NO DATA	NO DATA	NO DATA

Acute toxicity to terrestrial animals is categorized using the classification system shown in Table 23 (U.S. EPA, 2004). Toxicity categories for terrestrial plants have not been defined.

Table 23. Categories of Acute Toxicity for Avian and Mammalian Studies.

Toxicity Category	Oral LD_{50}	Dietary LC_{50}
Very highly toxic	< 10 mg/kg	< 50 ppm
Highly toxic	10 - 50 mg/kg	50 - 500 ppm
Moderately toxic	51 - 500 mg/kg	501 - 1000 ppm
Slightly toxic	501 - 2000 mg/kg	1001 - 5000 ppm
Practically non-toxic	> 2000 mg/kg	> 5000 ppm

4.2.1 Toxicity to Birds

As specified in the Overview Document, the Agency uses birds as a surrogate for terrestrial-phase amphibians when amphibian toxicity data are not available (U.S. EPA, 2004). No terrestrial-phase amphibian data are available for Ziram; therefore, acute and chronic avian toxicity data are used to assess the potential direct effects of Ziram to terrestrial-phase CRLFs.

4.2.1.1 Birds: Acute Exposure (Mortality) Studies

Results from the acute avian oral toxicity study with an LD50=97 mg/kg for the (*Colinus virginianus*) quail (MRID 417257-01) indicated that Ziram is moderately toxic to birds. This study was classified as acceptable and will be used in the CRLF assessment.

Results from an acute Ziram dietary toxicity study for (*Anas platyrhynchus*) mallard ducks (MRID 423863-002) resulted in an LC50=5156 mg/kg. This study was classified as acceptable. However, the acute oral toxicity value is more sensitive and will used in the assessment. A description of the study is found in Appendix C.

Toxicity Studies for Thiram are described in Appendix D. The risk estimates for the effect of Thiram to birds is provided as additional lines of evidence in the risk description section for two acute avian studies for the pheasant and the mallard duck Acute avian LD50s ranged from >100 mg/kg for the Red-wing Blackbird and Starling(MRID 075683) to >2800 mg/kg for the mallard (MRID BAOTH103).

4.2.1.2 Birds: Chronic Exposure (Growth, Reproduction) Studies

Toxicity Studies for the Major Degradate Thiram will be used in this assessment due to the persistence of the degradate. However, there is a Ziram reproduction study described in Appendix C. The one-generation reproductive toxicity of Thiram Technical to groups (16 pens/treatment level) of 1 male and 1 female of 17-week-old mallard ducks was assessed over approximately 23 weeks. Thiram was administered to the birds in the diet at measured concentrations of 0, 2.43, 9.61, and 39.7 mg ai/kg bw diet. There were significant reductions in several reproductive parameters at the highest treatment level, including eggs set, viable embryos, live 3-week embryos, normal hatchlings, 14-day old survivors, eggs set/eggs laid, normal hatchlings/live 3-week embryos, and normal hatchlings/eggs laid. As a result, the NOAEC was determined to be 9.6 mg/kg a.i. This study (MRID 45441202) is classified as acceptable and will be used in the assessment due to no open literature available with more sensitive endpoints.

4.2.1.3 Open Literature for Toxicity to Birds

Registrant submitted studies as well as open literature were reviewed for avian data. No avian toxicity data with more sensitive endpoints was available in ECOTOX..

4.2.2 Toxicity to Mammals

Mammalian toxicity data are used to assess potential indirect effects of Ziram to the terrestrial-phase CRLF. Effects to small mammals resulting from exposure to Ziram has the potential to also indirectly affect the CRLF via reduction in available food. As discussed in Section 2.5.3, over 50% of the prey mass of the CRLF may consist of vertebrates such as mice, frogs, and fish (Hayes and Tennant, 1985).

4.2.2.1 Mammals: Acute Exposure (Mortality) Studies

To estimate the indirect effect of Ziram on prey items, registrant submitted toxicity studies as well as open literature was reviewed for mammal data. An acute oral rat study (MRID 413404-01) resulted in an LD50=320 mg/kg (males). This indicates that Ziram is moderately toxic to mammals. This value will be used in calculations described in the risk estimation section due to no open literature available with a more sensitive endpoint..

In addition to reviewing toxicity studies for the parent Ziram, studies were also reviewed for the degradate Thiram. An acute oral rat study for Thiram resulted in an LD50= 2600 mg/kg (MRID 00153548) classifying Thiram as practically non-toxic to mammals. This study is described in Appendix D.

4.2.2.2 Mammals: Chronic Exposure (Growth, Reproduction) Studies

Although there is a reproductive rat study for Ziram (MRID 439358-01) resulting in a NOAEL=207 mg/kg), the chronic rat study for Thiram will be used in the risk estimation section due to the persistence. In addition to the persistence of Thiram, the reproductive study also indicated that Thiram is more sensitive based on a lower NOAEC. A Thiram rat reproductive study (MRID 420959-01) resulted in a NOAEC=1.9mg/kg.

4.2.2.3 Open Literature for Mammals

In addition to reviewing registrant submitted studies, open literature from ECOTOX was reviewed. Thiram rat reproductive values will be used to assess chronic effects of Ziram due to the persistence of the degradate Thiram. No Ziram open literature was available with more sensitive endpoints. No open literature available from the Thiram RED (09/30/04) s available with a more sensitive endpoint.

4.2.3 Toxicity to Terrestrial Invertebrates

Terrestrial invertebrate toxicity data are used to assess potential indirect effects of Ziram to the terrestrial-phase CRLF. Effects to terrestrial invertebrates resulting from exposure to Ziram has the potential to also indirectly affect the CRLF via reduction in available food.

4.2.3.1 Terrestrial Invertebrates: Acute Exposure (Mortality) Studies

Toxicity studies submitted to be used to assess the risk to terrestrial invertebrates include studies for both the parent Ziram and the degradate Thiram. A Ziram toxicity study for the honey bee (MRID 416679-01) was classified as acceptable. No negative control was used and the only treatment level was 100 µg/bee. This study resulted in no mortality for the solvent control and 6/100 mortalities (6%) in the 100 µg/bee treatment level. This study indicates that Ziram is practically nontoxic to bees based on the LD50>100 µg/bee.

There is also a toxicity value for Thiram (LD50=73.72 µg/bee) which indicates bees are more sensitive to Thiram. Due to the indeterminate endpoint for Ziram (LD50>100 µg/bee and the more sensitive Thiram endpoint, the toxicity value for Thiram will be used in the risk estimation section to support the determination.

4.2.3.2 Terrestrial Invertebrates: Open Literature Studies

A review of terrestrial invertebrate studies from the open literature was conducted. No open literature Ziram studies were available with more sensitive endpoints.

4.2.4 Toxicity to Terrestrial Plants

Terrestrial plant toxicity data are used to evaluate the potential for Ziram to affect riparian zone and upland vegetation within the action area for the CRLF. Impacts to riparian and upland (i.e., grassland, woodland) vegetation may result in indirect effects to both aquatic- and terrestrial-phase CRLFs, as well as modification to designated critical habitat PCEs via increased sedimentation, alteration in water quality, and reduction in of upland and riparian habitat that provides shelter, foraging, predator avoidance and dispersal for juvenile and adult CRLFs.

Plant toxicity data from both registrant-submitted studies and studies in the scientific literature were reviewed for this assessment. Registrant-submitted studies are conducted under conditions and with species defined in EPA toxicity test guidelines. Sub-lethal endpoints such as plant growth, dry weight, and biomass are evaluated for both monocots and dicots, and effects are evaluated at both seedling emergence and vegetative life stages. Guideline studies generally evaluate toxicity to ten crop species. A drawback to these tests is that they are conducted on herbaceous crop species only, and extrapolation of effects to other species, such as the woody shrubs and trees and wild herbaceous species, contributes uncertainty to risk conclusions.

Commercial crop species have been selectively bred, and may be more or less resistant to particular stressors than wild herbs and forbs. The direction of this uncertainty for specific plants and stressors, including Ziram, is largely unknown. Homogenous test plant seed lots also lack the genetic variation that occurs in natural populations, so the range of effects seen from tests is likely to be smaller than would be expected from wild populations.

A Seedling Emergence toxicity test (MRID 468931-01) for monocots resulted in an EC25 >6lbs/A NOAEC=6 lbs/A. No treatment effects were observed for the highest concentration. This study was classified as acceptable.

A Seedling Emergence toxicity test (MRID 468931-01) for dicots resulted in an EC25 >6lbs/A NOAEC=6 lbs/A. No treatment effects were observed for the highest concentration. Most sensitive dicot was soybean based on 16% reduction relative to negative control. This study was classified as acceptable.

A vegetative vigor toxicity test (MRID 468931-02) for monocots resulted in an EC25 >6lbs/A NOAEC<6 lbs/A. Most sensitive monocot was rye grass with a 13% reduction in comparison to the control. No treatment effects were observed for the highest concentration. This study was classified as acceptable.

A vegetative vigor toxicity test (MRID 468931-02) for dicots resulted in an EC25 >6lbs/A NOAEC=6 lbs/A. No treatment effects were observed for the highest concentration. Most sensitive dicot was tomato with a 12% reduction in comparison to the control. This study was classified as acceptable.

The results of the Tier II seedling emergence and vegetative vigor toxicity tests on non-target plants are summarized in 5.1.2.3**.**

Although toxicity data for terrestrial plants is available for the parent Ziram, no terrestrial plant studies are available for Thiram. There are no open literature studies with more sensitive endpoints for Ziram available. Therefore risk to terrestrial plants for this assessment are based on Ziram toxicity values.

4.2.5 Terrestrial Plant Open Literature

In addition to reviewing the registrant submitted studies for terrestrial plant data for Ziram, a search of open literature in the ECOTOX database was conducted. No terrestrial plant studies resulted in more sensitive endpoints, so the registrant submitted data was used to assess the risk to terrestrial plants for effects on indirect critical habitat .

4.2.6 Sublethal Effects

In addition to the rat reproduction studies, studies were also submitted to determine the carcinogenic effects of Ziram. Results from studies indicate Ziram has carcinogenic effects. The CARC concluded that Ziram showed evidence of carcinogenicity based on two studies. One study (MRID 434042-01) indicated Ziram was carcinogenic to male rats and one study indicated effects on female rats (MRID 457702-01434042-01):

! In a 1994 study with the agricultural grade Ziram (98.7% a.i.) (MRID # 43404201), male CD (SD) BR rats had a significant positive trend for mesenteric lymph node hemangiomas and combined incidences of mesenteric lymph node and spleen hemangiomas. There was also a significant increase by pair-wise comparison with the controls for mesenteric lymph node hemangiomas and combined incidence of mesenteric lymph node and spleen hemangiomas at the highest dose tested of 540 ppm (23.7 mg/kg/day). The incidences of these tumors were outside the historical controls range (0%-4%). The CARC, therefore, concluded that these benign tumors in males were treatment-related. There were no compound-related tumors observed in female CD (SD) BR rats. The statistical evaluation of mortality indicated no significant incremental changes with increasing doses of Ziram in male rats. Female rats showed a significant decreasing trend in mortality with increasing doses of Ziram.

In a 1983 study with agricultural grade Ziram (97.5% a.i.) (MRID # 45770201), male F344 rats had a significant increasing trend in preputial gland adenomas, but there was no significant difference in pair-wise comparisons of the dosed groups with controls. Additionally, glandular hyperplasia was increased at the high dose greatly in excess of the historical control incidence. The CARC, therefore, concluded that these benign tumors in males were suggestive of a treatment-related response. No carcinomas were observed. There were no compound related tumors observed in female F344 v rats. The dosing at 2000 ppm was considered to be adequate and not excessive by the Committee. This was based on a significant trend for increased mortality with dose in males, decreased body weight gain and food consumption in both sexes, clinical pathology findings, organ weight changes including a decrease in absolute brain weight in both sexes, and increased histopathological changes in various organs, none of which were considered to be severely adverse.

4.3 Use of Probit Slope Response Relationship to Provide Information on the Endangered Species Levels of Concern

The Agency uses the probit dose response relationship as a tool for providing additional information on the potential for acute direct effects to individual listed species and aquatic animals that may indirectly affect the listed species of concern (U.S. EPA, 2004). As part of the risk characterization, an interpretation of acute RQ for listed species is discussed. This interpretation is presented in terms of the chance of an individual event (i.e., mortality or immobilization) should exposure at the EEC actually occur for a species with sensitivity to Ziram on par with the acute toxicity endpoint selected for RQ calculation. To accomplish this interpretation, the Agency uses the slope of the dose response relationship available from the toxicity study used to establish the acute toxicity measures of effect for each taxonomic group that is relevant to this assessment. The individual effects probability associated with the acute RQ is based on the mean estimate of the slope and an assumption of a probit dose response relationship. In addition to a single effects probability estimate based on the mean, upper and lower estimates of the effects probability are also provided to account for variance in the slope, if available.

Individual effect probabilities are calculated based on an Excel spreadsheet tool IECV1.1 (Individual Effect Chance Model Version 1.1) developed by the U.S. EPA, OPP, Environmental Fate and Effects Division (June 22, 2004). The model allows for such calculations by entering the mean slope estimate (and the 95% confidence bounds of that estimate) as the slope parameter for the spreadsheet. In addition, the acute RQ is entered as the desired threshold.

4.4 Incident Database Review

A review of the EIIS database for ecological incidents involving Ziram was completed on June 20, 2008. The results of this review for terrestrial, plant, and aquatic incidents are discussed below in Sections 4.4.1 through 4.4.3, respectively.

4.4.1 Terrestrial Incidents

Although there were no Ziram terrestrial incidents reported in the EIIS database, there was one terrestrial incident reported for Thiram. A report (I005754-012) from Bakersfield, California of 200 dead cedar waxwings was reported for Thiram. This report was classified as unlikely based on no pesticide residue detected in samples.

4.4.2 Plant Incidents

A report (I013563-02) of deformation in apricots was reported for Ziram. A registered use on March 23, 1999 covering 40 acres in San Joaquin, California was classified as possible. There were no plant incidents reported for Thiram.

4.4.3 Aquatic Incidents

No aquatic incidents were reported for Ziram in the EISS database. There were also no aquatic incidents reported for Thiram.

5. Risk Characterization

5.1 Risk Estimation

Risk is estimated by calculating the ratio of exposure to toxicity. This ratio is the risk quotient (RQ), which is then compared to pre-established acute and chronic levels of concern (LOCs) for each category evaluated (Appendix E). For acute exposures to the CRLF and its animal prey in aquatic habitats, as well as terrestrial invertebrates, the LOC is 0.05. For acute exposures to the CRLF and mammals, the LOC is 0.1. The LOC for chronic exposures to CRLF and its prey, as well as acute exposures to plants is 1.0.

Risk to the aquatic-phase CRLF is estimated by calculating the ratio of exposure to toxicity using 1-in-10 year EECs based on the label-recommended Ziram usage scenarios summarized in Table 15 and the appropriate aquatic toxicity endpoint from Table 11. Risks to the terrestrial-phase CRLF and its prey (*e.g.* terrestrial insects, small mammals and terrestrial-phase frogs) are estimated based on exposures resulting from applications of Ziram. Exposures are also derived for terrestrial plants, as discussed in Section 3.3 and the effects are summarized in Section 4.

5.1.1 Exposures in the Aquatic Habitat

5.1.1.1 Direct Effects to Aquatic-Phase CRLF

Direct effects to the aquatic-phase CRLF are based on peak EECs in the standard pond and the lowest acute toxicity value for freshwater fish. In order to assess direct chronic risks to the CRLF, 60-day EECs and the lowest chronic toxicity value for freshwater fish are used. Ziram is likely to directly affect the aquatic-phase of the CRLF.

Acute Ziram Effects

To determine the direct acute effects of Ziram on the CRLF, RQs were estimated using the peak EECs and the Ziram toxicity values. RQs for Ziram range from 1.24 for cherries to 5.46 for wine grapes. There is a "may affect" determination for Ziram based on RQs for all use scenarios exceeding the LOC=0.05 for acute effects.

In addition to determining if the acute RQ exceeds the endangered species LOC, the probit analysis is used to refine the risk. The probit analysis estimates the chance of individual effects. Using the slope=6.15, the chance of individual effects ranges from about 1 in 1.39 for cherries with a ground application to about 1 in 1.00 (almonds applied aerially, apricots, nectarines, peaches, pecans, and wine grapes).

Acute Thiram Effects

Acute RQs for Thiram range from 0.70 for pears to 5.89 for nectarines. There is a "may affect" determination for Thiram based on RQs for all use scenarios exceeding the LOC=0.05.

The probit analysis for Thiram, using the default slope of 4.5 indicates the chance of individual effects ranges from about 1 in 4.12 for pears to about 1 in 1.00 for nectarines, peaches, and wine grapes.

Table 24. Summary of Direct Effect RQs[a] for the Aquatic-phase CRLF.

Use Scenario	Surrogate Species	Toxicity Value (µg/L)	EEC (µg/L)[b]	RQ	Probability of Individual Effect at ES LOC[c]	Probability of Individual Effect at RQ[c]
Acute Direct Toxicity Using Ziram Toxicity Values						
Almonds Ground	Fathead Minnow	LC_{50} = 8.07	Peak: 16.75	**2.09**	1 in 1.62E+15	**1 in 1.03**
Almonds aerial			16.75	**2.93**		**1 in 1.00**
Apples			15.14	**1.89**		**1 in 1.05**
Apricots ground			23.51	**2.94**		**1 in 1.00**
Apricots aerial			32.94	**4.12**		**1 in 1.00**

Cherries ground			9.89	**1.24**		**1 in 1.39**
Cherries aerial			13.31	**1.66**		**1 in 1.10**
Grapes ground			13.47	**1.68**		**1 in 1.09**
Grapes aerial			15.82	**1.98**		**1 in 1.04**
Nectarines ground			34.61	**4.33**		**1 in 1.00**
Nectarines aerial			42.16	**5.27**		**1 in 1.00**
Peaches ground			32.58	**4.07**		**1 in 1.00**
Peaches aerial			35.64	**4.46**		**1 in 1.00**
Pears			14.94	**1.87**		**1 in 1.05**
Pecans			28.63	**3.58**		**1 in 1.00**
Wine Grapes ground			43.70	**5.46**		**1 in 1.00**
Wine Grapes-aerial			42.46	**5.31**		**1 in 1.00**
Acute Direct Toxicity of Thiram (Using Default Slope=4.5 for Probit Analysis)						
Almonds	Harlequinn	LC_{50} = 7	**25.64**	**3.66**	1 in 4.18E+08	**1 in 1.01**
Apples			**17.23**	**2.46**		**1 in 1.04**
Apricots			**25.98**	**3.71**		**1 in 1.01**
Cherries			**15.26**	**2.18**		**1 in 1.07**
Grapes			**12.59**	**1.80**		**1 in 1.14**
Nectarines			**41.26**	**5.89**		**1 in 1.00**
Peaches			**30.00**	**4.28**		**1 in 1.00**
Pears			**4.88**	**0.70**		**1 in 4.12**
Pecans			**15.82**	**2.26**		**1 in 1.06**
Wine Grapes-aerial			**29.06**	**4.15**		**1 in 1.00**
Chronic Direct Effects Using Chronic Thiram Toxicity Value						
Almonds	Fathead minnow	NOAEC = 530.2	60 day 7.53	0.01	Not calculated for chronic endpoints	
Apples			3.37	0.01		
Apricots			7.22	0.01		
Cherries			4.29	0.01		
Grapes			5.36	0.01		
Nectarines			16.35	0.03		
Peaches			10.86	0.02		
Pears			1.25	0.01		
Pecans			3.59	0.01		
Wine Grapes-aerial			8.53	0.02		

[a]RQs associated with acute and chronic direct toxicity to the CRLF are also used to assess potential indirect effects to the CRLF based on a reduction in freshwater fish and frogs as food items.
[b]The highest EEC based on Ziram use on peaches at 7.6 lb ai/A/acre/year (see Table 3.3).
[c]The probit slope value for the Ziram acute bluegill sunfish toxicity test is 6.15
Bold: Exceeds Acute endangered species LOC of 0.05. for aquatic effects

In addition to estimating the acute risk of Ziram on the aquatic-phase CRLF using the fish as a surrogate, the chronic risk is also estimated. Due to the persistence of the degradate Thiram, the toxicity values for Thiram will be used to estimate chronic risk. However, due to the absence of

Thiram chronic data for fish, the acute-to-chronic value = 530.2 µg/L for Thiram is used. RQs range from 0.01 for almonds, apples, apricots, cherries, grapes, and pecans to 0.03 for nectarines using the ACR value. A "no effect" determination is based on no LOC exceedence for chronic effects for fish as a surrogate for the CRLF.

5.1.1.2 Indirect Effects to Aquatic-Phase CRLF via Reduction in Prey (non-vascular aquatic plants, aquatic invertebrates, fish, and frogs)

Non-vascular Aquatic Plants

Indirect effects of Ziram to the aquatic-phase CRLF (tadpoles) via reduction in non-vascular aquatic plants for dietary and habitat effects are based on peak EECs from the standard pond and the lowest acute toxicity value for aquatic non-vascular plants (*Pseudokirchneriella subcapitata* EC_{50}=67 µg/L for Ziram. Based on all use scenarios resulting in no LOC exceedence, there is a "no effect" determination for Ziram for the CRLF via reduction in non-vascular plants (Table 25).

Table 25. Summary of Acute RQs Used to Estimate Indirect Effects to the CRLF via Effects to Non-Vascular Aquatic Plants (diet of CRLF in tadpole life stage and habitat of aquatic-phase CRLF).

Ziram (EC50=67 ppb)			
Uses	**Application rate (lb ai/A) and type**	**Peak EEC (µg/L)**	**Indirect effects RQ* (food and habitat)**
Almonds	6.1	16.75	025
Almonds aerial	6.1	23.44	0.35
Apples	4.6	15.14	0.23
Apricots-ground	6.1	23.51	035
Apricots aerial	6.1	32.94	0.49
Cherries ground	3.8	9.89	0.15
Cherries aerial	3.8	13.31	0.20
Grapes-aerial	3.0	15.82	0.24
Nectarines-ground	7.6	34.61	0.52
Nectarines-aerial	7.6	42.16	0.63
Peaches-ground	7.6	32.58	0.49
Peaches aerial	7.6	35.64	0.53
Pears	4.6	14.94	0.22
Pecans	6.1	28.63	0.43
Wine Grapes	3.0	42.46	0.63

	Aerial		
Thiram EC50=140 ppb)			
Uses	**Application rate (lb ai/A) and type**	**Peak EEC (μg/L)**	**Indirect effects RQ* (food and habitat)**
Almonds	6.1	25.64	0.18
Apples	4.6	17.23	0.12
Apricots	6.1	25.98	0.18
Cherries	3.8	15.26	0.11
Grapes	3.0	12.59	0.09
Nectarines	7.6	41.26	0.29
Peaches	7.6	30.00	0.21
Pears	**4.6**	4.88	0.03
Pecans	6.1	15.82	0.11
Wine Grapes-aerial	3.0 aerial	29.06	0.21
[1] LOC exceedances (RQ ≥ 1) are bolded and shaded. RQ = use-specific peak EEC/ [insert most sensitive non-vascular aquatic plant endpoint here].			

Aquatic Invertebrates

Indirect acute effects to the aquatic-phase CRLF via effects to prey (invertebrates) in aquatic habitats are based on peak EECs in the standard pond and the lowest acute toxicity value for freshwater invertebrates. For chronic risks, 21-day EECs and the lowest chronic toxicity value for invertebrates are used to derive RQs. A summary of the acute and chronic RQ values for exposure to aquatic invertebrates (as prey items of aquatic-phase CRLFs) is provided in Table 26. Based on the RQ exceedence for all use scenarios for acute effects, Ziram results in a "may affect" determination for the CRLF via reduction in freshwater invertebrates prey items.

Table 26. Summary of Acute and Chronic RQs Used to Estimate Indirect Effects to the CRLF Via Direct Effects on Aquatic Invertebrates as Dietary Food Items (Prey of CRLF Juveniles and Adults in Aquatic Habitats).

Use Scenario	**Surrogate Species**	**Toxicity Value (μg/L)**	**EEC (μg/L)**	**RQ[1]**	**Probability of Individual Effect at ES LOC[2]**	**Probability of Individual Effect at RQ[2]**
Acute Indirect Toxicity for Ziram (Peak)						
Almonds	*Daphnia*	LC_{50} = 48	16.75	**0.35**	1 in 208	1 in 5.49
Apples			14.94	**0.31**		1 in 6.42
Apricots			23.51	**0.49**		1 in 3.72
Cherries			9.89	**0.21**		1 in 11.3
Grapes			12.62	**0.26**		1 in 8.19
Nectarines			34.61	**0.72**		1 in 2.58
Peaches			32.58	**0.68**		1 in 2.71
Pears			28.63	**0.60**		1 in 3.04

Use Scenario	Surrogate Species	Toxicity Value (µg/L)	EEC (µg/L)	RQ[1]	Probability of Individual Effect at ES LOC[2]	Probability of Individual Effect at RQ[2]
Wine Grapes Ground			43.70	**0.91**		1 in 2.14
Wine Grapes Aerial			42.46	**0.88**		1 in 2.19
Chronic Direct Toxicity (21 Day)						
Almonds	*Daphnia*	NOAEC = 170.6	14.47	0.04	Not calculated for chronic endpoints	
Apples			8.49	0.03		
Apricots			14.06	0.05		
Cherries			8.62	0.02		
Grapes			6.67	0.02		
Nectarines			24.38	0.08		
Peaches			16.13	0.01		
Pears			2.32	0.01		
Pecans			5.72	0.1		
Wine Grapes Aerial			13.69	0.2		

[1]RQs associated with acute and chronic direct toxicity to the CRLF are also used to assess potential indirect effects to the CRLF based on a reduction in freshwater fish and frogs as food items.
[2]The probit slope value for the acute daphnia toxicity test is 1.99.
Bold: Exceeds Acute endangered species LOC of 0.05 and LOC of 1 for chronic effects.

In addition to estimating the effects of Ziram on acute indirect prey reduction, the chronic effect on prey reduction of Ziram on the aquatic-phase CRLF is also calculated. Due to the persistence of the degradate Thiram, the toxicity values for Thiram will be used to estimate chronic risk. However, due to the absence of Thiram chronic data for *Daphnia*, the acute-to-chronic value = 170.6 µg/L for Thiram is used. RQs range from 0.01 for pears, to 0.14 for nectarines using the ACR value. A “no effect” determination is based on no LOC exceedence for chronic effects for *Daphnia* as prey for the CRLF.

Fish and Frogs

Fish and frogs also represent potential prey items of adult aquatic-phase CRLFs. RQs associated with acute and chronic direct toxicity to the CRLF (Table 24) are used to assess potential indirect effects to the CRLF based on a reduction in freshwater fish and frogs as food items. There are no data available to calculate RQs for frogs therefore risk is assessed using fish as a surrogate for the frog.. Based on the LOC exceedences for all use scenarios using fish as prey and fish as surrogates for frogs , Ziram resulted in a “may affect” determination for the CRLF via reduction in freshwater fish and frogs as food items.

5.1.1.3 Indirect Effects to CRLF via Reduction in Habitat and/or Primary Productivity (Freshwater Aquatic Plants)

Indirect effects to the CRLF via direct toxicity to aquatic plants are estimated using the most sensitive non-vascular and vascular plant toxicity endpoints. Because there are no obligate relationships between the CRLF and any aquatic plant species, the most sensitive EC_{50} values, rather than NOAEC values, were used to derive RQs. Table 27 includes RQs for vascular plants and RQs for non-vascular plants. There is a "No Effect" determination for indirect effects to the CRLF via reduction in vascular plants based on no LOC exceedences for any use scenarios.

Table 27. Summary of Acute RQs Used to Estimate Indirect Effects to the CRLF via Effects to Vascular Aquatic Plants (habitat of aquatic-phase CRLF)a

Uses	Application rate (lb ai/A) and type	Peak EEC (µg/L)	Indirect effects RQ[1] (food and habitat)
Almonds	6.1	16.75	0.04
Apples/Pears	4.6	14.94	0.04
Apricots	6.1	23.51	0.07
Cherries	3.8	9.89	0.03
Grapes	3.0	12.62	0.34
Peaches	7.6	32.58	0.09
Nectarines	7.6	34.61	0.09
Pecans	6.1	28.63	0.08
Wine grapes Ground	3.0 ground	43.70	0.12
Wine Grapes Aerial	3.0 aerial	42.46	0.11

[1] RQs used to estimate indirect effects to the CRLF via toxicity to non-vascular aquatic plants are summarized in Table 25.
Bold: LOC exceedances (RQ $\geq$ 1)a. RQ = use-specific peak EEC / 370 µg/L.

5.1.2 Exposures in the Terrestrial Habitat

5.1.2.1 Direct Effects to Terrestrial-phase CRLF

As previously discussed in Section 3.3, potential direct effects to terrestrial-phase CRLFs are based on foliar applications of Ziram.

Potential direct acute effects to the terrestrial-phase CRLF are derived by considering dose- and dietary-based EECs modeled in T-REX for a small bird (20 g) consuming small invertebrates (Table 28) and acute oral and subacute dietary toxicity endpoints for avian species.

Based on the LOC exceedence for all use scenarios (almonds, apples/pears, apricots, cherries, grapes, nectarines/peaches and pecans), RQs for acute effects range from 30.66 for cherries to 86.93 for peaches. Further analysis using the probit model resulted in all use scenarios having a 1 in 1.00 chance of individual effects due to Ziram.

Table 28. Summary of Direct Effect RQs for the Terrestrial-phase CRLF.

Use Scenario	Surrogate Species	Toxicity Value (mg/L)	EEC (mg/L)[1]	RQ[2]	Probability of Individual Effect at ES LOC[3]	Probability of Individual Effect at RQ[3]
Acute Direct Toxicity LD50 (based on Ziram)						
Almonds	Bird	97	3439	**49.2**	1 in 1.00	1 in 1.00
Apples/Pears			2594	**37.1**		1 in 1.00
Apricots			3439	**49.2**		1 in 1.00
Cherries			2142	**30.7**		1 in 1.00
All Grapes			2213	**31.7**		1 in 1.00
Nectarines/Peaches			6075	**86.9**		1 in 1.00
Pecans			2529	**36.2**		1 in 1.00
Chronic Direct Toxicity NOAEC (based on Thiram)						
Almonds	Bird	9.6	3020	**104.1**	Not calculated for chronic endpoints	
Apples/Pears			2277	**78.5**		
Apricots			3020	**104.1**		
Cherries			1881	**64.9**		
All Grapes			1943	**67.0**		
Nectarines/Peaches			5334	**183.9**		
Pecans			2221	**76.6**		

[1]The highest EEC based on Ziram use on peaches at 7.6 lb ai/A/acre/year.
[2]RQs associated with acute and chronic direct toxicity to the CRLF are also used to assess potential indirect effects to the CRLF based on a reduction in freshwater fish and frogs as food.
[3] The probit slope value for the acute quail toxicity test is the default slope=4.5.
Bold: Exceeds Acute endangered species LOC of 0.1

Potential direct chronic effects of Ziram to the terrestrial-phase CRLF are derived by considering dietary-based exposures modeled in T-REX for a small bird (20g) consuming small invertebrates. Chronic effects are estimated using the lowest available toxicity data for birds. EECs are divided by toxicity values to estimate chronic dietary-based RQs.
Thiram RQs for chronic effects range from for 64.87 cherries to 183.92 for nectarines/peaches. Ziram is likely to directly affect the terrestrial-phase of the CRLF.

5.1.2.2 Indirect Effects to Terrestrial-Phase CRLF via Reduction in Prey (terrestrial invertebrates, mammals, and frogs)

5.1.2.2.1 Terrestrial Invertebrates

In order to assess the risks of Ziram to terrestrial invertebrates, which are considered prey of CRLF in terrestrial habitats, the honey bee is used as a surrogate for terrestrial invertebrates. The toxicity value for terrestrial invertebrates is calculated by multiplying the acute contact LD_{50} by 1 bee/0.128g, which is based on the weight of an adult honey bee. EECs (μg a.i./g of bee) calculated by T-REX for small and large insects are divided by the calculated toxicity value for terrestrial invertebrates.

RQs were estimated for small and large insects to assess the effect of Ziram on indirect effects via reduction in prey. The Ziram LD50>100 µg/bee resulted in a "May Affect" determination based on the upper-bound RQs exceeding the LOC=0.5 for all uses for small and large insects. No probit analysis was conducted for Ziram due to the indeterminate endpoint.

An "LAA" determination for indirect effects of prey reduction results from RQs exceeding the acute risk LOC=0.5. An "LAA' determination resulted from the RQs exceeding the LOC=0.5 for all uses small insects. Large insect RQs exceed the acute risk LOC=0.5 for nectarines/peaches and pecans.

For those large insect RQs ranging between the endangered species LOC=0.05 and the acute risk LOC=0.5 (almonds, apples/pears, apricots, cherries and grapes) further analysis is provided. Due to the indeterminate Ziram endpoint, probit is not used to refine the analysis, but additional analysis based on the Thiram LD50=73.72is provided. Small insect RQs resulting from the T-REX model indicated potential effects of Thiram based on LOC (0.05) exceedences for all use scenarios. RQs for small insects ranged from 3.27 for cherries to 9.26 for peaches. RQs for large insects ranged from 0.36 for cherries to 1.03 for peaches. The results based on Thiram RQs for all uses for small insect RQs exceeding the LOC=0.05 supports the "May Affect" determination. The "LAA' determination is supported for indirect effects of terrestrial invertebrates as prey consumed by the CRLF is based on the all uses exceeding the acute risk LOC=0.5 for small insects.

The results for the probit refinement for Thiram supports the "LAA" determination for the CRLF consuming large insects for all uses. The chance of individual effects ranging from about 1 in 1.98 for nectarines/peaches to 3.33 for cherries for large insects.
d t Table 29 provides the results for each modeled use.

Table 29. Summary of RQs Used to Estimate Indirect Effects to the Terrestrial-phase CRLF Via Direct Effects on Terrestrial Invertebrates as Dietary Food Items Based on the Degradate Thiram as the Most Sensitive Toxicity Value.

Use Scenario	**Species**	**Toxicity Value (µg/L)**	**Small Insect EEC (µg/L)[b]**	**RQ[a]**	**Large Insect EECs**	**RQs[a]**	**Probability of Individual Effect at RQ for Small Insects[c]**	**Probability of Individual Effect at RQ for large Insects[c]**
Acute Indirect Toxicity for the Effects of Ziram on the CRLF Consuming Terrestrial Invertebrates								
Almonds		**LD50> 100**	3391	**<4.34**	377	**<0.48**		
Apples/ pears			2245	**<2.87**	249	**<0.32**		
Apricots			3391	**<4.34**	377	**<0.48**		
Cherries			1464	**<1.87**	163	**<0.21**		
Grapes			20582	**<2.63**	229	**<0.29**		
Peaches			5213	**<6.67**	579	**<0.74**		
Pecans			4184	**<5.35**	465	**<0.59**		
Acute Indirect Toxicity for the Effects of Thiram on the CRLF Consuming Terrestrial Invertebrates								

Use Scenario	Species	Toxicity Value (µg/L)	Small Insect EEC (µg/L)[b]	RQ[a]	Large Insect EECs	RQs[a]	Probability of Individual Effect at RQ for Small Insects[c]	Probability of Individual Effect at RQ for large Insects[c]
						Probability of Individual Effect at LOC		**About 1 in 16**
Almonds	Bee	LD50=73.72	3020	**5.24**		**0.58**	**About 1 in 1.25**	**About 1 in 2.56**
Apples/ Pears			2277	**3.95**		**0.44**	**About 1 in 1.32**	**About 1 in 2.97**
Apricots			3020	**5.24**		**0.58**	**About 1 in 1.25**	**About 1 in 2.56**
Cherries			1881	**3.27**		**0.36**	**About 1 in 1.37**	**About 1 in 3.33**
Grapes			1943	**3.37**		**0.37**	**About 1 in 1.36**	**About 1 in 3.28**
Peaches			5334	**9.26**		**1.03**	**About 1 in 1.15**	**About 1 in 1.98**
Pecans			2221	**3.86**		**0.43**	**About 1 in 1.32**	**About 1 in 3.01**

[a]RQs associated with acute a direct toxicity to the CRLF are also used to assess potential indirect effects to the CRLF based on a reduction in terrestrial invertebrates (honey bee) as food.
[b]The highest EEC based on peaches at 7.6lb ai/A/acre/year.
[c] The probit slope value is not available for the acute honey bee toxicity test.
Exceedeence of acute endangered species LOC of 0.05 in bold.

5.1.2.2.2 Mammals

Risks associated with ingestion of small mammals by large terrestrial-phase CRLFs are derived for dietary-based and dose-based exposures modeled in T-REX for a small mammal (15g) consuming short grass. Acute and chronic effects are estimated using the most sensitive mammalian toxicity data. EECs are divided by the toxicity value to estimate acute and chronic dose-based RQs as well as chronic dietary-based RQs. Based on LOC exceedences for all use scenarios (almonds, apples/pears, apricots, cherries, grapes, peaches and pecans) for both acute and chronic effects on a small mammal consuming short grass listed in Table 30. Ziram is likely to indirectly affect the CRLF via reduction in small mammal prey items. RQs for acute effects range from 4.53 for cherries to 12.85 for peaches. Ziram RQs for chronic dose-based effects range from140.17 for cherries to 397.42 for nectarines/peaches. RQs for chronic dietary-based effects range from for 16.16 for cherries to 45.81 for peaches. Thiram RQs for acute effects range from 0.43 for cherries to 1.55 for nectarines/peaches. RQs for chronic dose-based effects range from for 11888.01 for cherries to 42317.99 for nectarines/peaches. RQs for chronic dietary-based effects range from for 1370.21 for cherries to 4877.57 for nectarines/peaches.

Table 30. Summary of Acute and Chronic RQs Used to Estimate Indirect Effects to the Terrestrial-phase CRLF via Direct Effects on Small Mammals as Dietary Food Items.

Ziram			
Use (Application Rate)	**Chronic RQ**		**Acute RQ**
	Dose-based Chronic RQ[1]	**Dietary-based Chronic RQ[2]**	**Dose-based Acute RQ[3]**
Almonds	**225.01**	**25.93**	**7.28**
Apples/Pears	**169.68**	**19.56**	**5.49**
Apricots	**225.01**	**25.93**	**7.28**
Cherries	**140.17**	**16.16**	**4.53**
All Grapes	**144.79**	**16.69**	**4.68**
Peaches	**397.42**	**45.81**	**12.85**
Pecans	**165.47**	**19.07**	**5.35**

Bold: LOC exceedances (acute RQ $\geq$ 0.1 and chronic RQ $\geq$ 1) are bolded and shaded.
[1] Based on dose-based EEC and Ziram rat NOAEL = 10.35 mg/kg-bw.
[2] Based on dietary-based EEC and Ziram rat NOAEC = 207 mg/kg-diet.
[3] Based on dose-based EEC and Ziram rat acute oral LD_{50} = 320 mg/kg-bw.

Thiram			
Use (Application Rate)	**Chronic RQ**		Acute RQ
	Dose-based Chronic RQ[1]	**Dietary-based Chronic RQ[2]**	**Dose-based Acute RQ[3]**
Almonds	**24172**	**2786**	**0.88**
Apples/Pears	**18228**	**2101**	**0.67**
Apricots	**24172**	**2786**	**0.88**
Cherries	**11888**	**1370**	**0.43**
All Grapes	**16704**	**1825**	**0.61**
Nectarines/Peaches	**42318**	**4877**	**1.55**
Pecans	**33966**	**3915**	**1.24**

Bold: LOC exceedances (acute RQ $\geq$ 0.1 and chronic RQ $\geq$ 1) are bolded and shaded.
[1] Based on dose-based EEC and Thiram rat NOAEL = 0.10 mg/kg-bw.
[2] Based on dietary-based EEC and Thiram rat NOAEC = 1.90 mg/kg-diet.
[3] Based on dose-based EEC and Ziram rat acute oral LD_{50} = 2600 mg/kg-bw.

5.1.2.2.3 Frogs

An additional prey item of the adult terrestrial-phase CRLF is other species of frogs. In order to assess risks to these organisms, dietary-based and dose-based exposures modeled in T-REX for a small bird (20g) consuming small invertebrates are used (See Section 5.1.2.1 and Table 28) for results. Acute Ziram RQs using birds as a surrogate for the terrestrial-phase CRLF ranged from 30.7 for cherries to 86.9 for nectarines/peaches. An “LAA” determination for indirect effects via reduction in prey is based on the RQs using the bird as a surrogate for frogs exceeding the acute risk LOC=0.5 for all uses. Additional support for the “LAA” determination for acute effects is provided by the probit results. All uses indicate about a 1 in 1 chance of individual effect.

To provide support for the “LAA” determination chronic Thiram RQs were also calculated. RQs for chronic effects range from 64.9 for cherries to 183.9 for nectarines/peaches.

5.1.2.3 Indirect Effects to CRLF via Reduction in Terrestrial Plant Community (Riparian and Upland Habitat)

Potential indirect effects to the CRLF resulting from direct effects on riparian and upland vegetation are assessed using RQs from terrestrial plant seedling emergence and vegetative vigor EC_{25} data as a screen.

Although the maximum label rate is 7.6 lb/A for peaches, the terrestrial plant studies were conducted with one concentration of 6.1 lb/A, near the label rate. No EC50 was estimated from the toxicity studies due to no treatment-related effects at the highest tested concentration observed for either the seedling emergence or vegetative vigor studies. Based on the results of the seedling emergence and vegetative studies, no RQs will be estimated using the TerrPlant model. Therefore, there are no indirect effects to the CRLF or its habitat from effects to riparian or upland plants.

5.1.3 Primary Constituent Elements of Designated Critical Habitat

For Ziram use, the assessment endpoints for designated critical habitat PCEs involve a reduction and/or modification of food sources necessary for normal growth and viability of aquatic-phase CRLFs, and/or a reduction and/or modification of food sources for terrestrial-phase juveniles and adults. Because these endpoints are also being assessed relative to the potential for indirect effects to aquatic- and terrestrial-phase CRLF, the “Likely to Adversely Affect” determination for the aquatic-phase CRLF based on aquatic invertebrates and for the terrestrial-phase CRLF based on fish and frogs, terrestrial invertebrates and small mammals for indirect effects from the potential loss of food items are used as the basis of the effects determination for potential modification to designated critical habitat.

5.1.3.1 Aquatic-Phase (Aquatic Breeding Habitat and Aquatic Non-Breeding Habitat)

Three of the four assessment endpoints for the aquatic-phase primary constituent elements (PCEs) of designated critical habitat for the CRLF are related to potential effects to aquatic and/or terrestrial plants:

- Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.

- Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.
- Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.,* algae).

Based on the risk estimation for potential effects to aquatic and/or terrestrial plants provided in Sections 5.1.1.2, 5.1.1.3, and 5.1.2.3, Ziram will have no effect on aquatic-phase PCEs of designated habitat related to effects on aquatic and/or terrestrial plants. There is no LOC exceedence for aquatic vascular or non-vascular plants for Ziram. RQs for vascular plants range from 0.3 for grapes to 0.11 wine grapes with an aerial application.

The lines of evidence for riparian vegetation based on terrestrial plants indicate there is no potential for adverse effects as well. There was a "No Effect" determination based on no treatment related effects at the highest Ziram concentration tested.
The remaining aquatic-phase PCE is "alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source." To assess the impact of Ziram on this PCE, acute and chronic freshwater fish and invertebrate toxicity endpoints, as well endpoints for aquatic non-vascular plants, are used as measures of effects. RQs for these endpoints were calculated in Sections 5.1.1.1 and 5.1.1.2. Although there were no LOC exceedences for any modeled crop for non-vascular plants, there were LOC exceedences for all use scenarios for both acute fish and aquatic invertebrates. Based on LOC exceedences for all use scenarios for acute fish and acute aquatic invertebrates, Ziram results in habitat modification relative to the aquatic-phase PCE regarding alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.

5.1.3.2 Terrestrial-Phase (Upland Habitat and Dispersal Habitat)

Two of the four assessment endpoints for the terrestrial-phase PCEs of designated critical habitat for the CRLF are related to potential effects to terrestrial plants:

- Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or dripline surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance
- Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal

The risk estimation for terrestrial-phase PCEs of designated habitat related to potential effects on terrestrial plants is provided in Section 5.1.2.3. These results will inform the effects determination for modification of designated critical habitat for the CRLF.

The third terrestrial-phase PCE is "reduction and/or modification of food sources for terrestrial phase juveniles and adults." To assess the impact of Ziram on this PCE, acute and chronic toxicity endpoints for birds, mammals, and terrestrial invertebrates are used as measures of effects. RQs for these endpoints were calculated in Section 5.1.2.2. There is the potential for reduction or modification of food sources for the CRLF based on RQ exceedences and probit analysis for frogs, terrestrial invertebrates (bees) and small mammals. Therefore, Ziram results in a determination of habitat modification relative to this terrestrial-phase PCE.

The fourth terrestrial-phase PC is based on alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source. Direct acute and chronic RQs for terrestrial-phase CRLFs are presented in Section 5.2.1.2. There is a potential for alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLF based on RQ exceedences and probit analysis for aquatic invertebrates. There is the potential for normal growth and viability of juvenile and adult CRLF based on RQ exceedences and probit analysis for frogs, terrestrial invertebrates (bees) and small mammals. Due to the indirect dietary LOC exceedences for the CRLF consuming terrestrial invertebrates, mammals and frogs it is determined that Ziram results in habitat modification relative to this terrestrial-phase PCE.

5.2 Risk Description

The risk description synthesizes an overall conclusion regarding the likelihood of adverse impacts leading to an effects determination (*i.e.*, "no effect," "may affect, but not likely to adversely affect," or "likely to adversely affect") for the CRLF and its designated critical habitat.

If the RQs presented in the Risk Estimation (Section 5.1) show no direct or indirect effects for the CRLF, and no modification to PCEs of the CRLF's designated critical habitat, a "no effect" determination is made, based on Ziram's use within the action area. However, if direct or indirect effect LOCs are exceeded or effects may modify the PCEs of the CRLF's critical habitat, the Agency concludes a preliminary "may affect" determination for the FIFRA regulatory action regarding Ziram. Based on the LOC exceedences for direct and indirect effects for the aquatic and terrestrial-phase CRLF, there is a "may affect" determination. Based on the effects of Ziram on the PCEs for a reduction in resources, there is a habitat modification determination for critical habitat. There is also a habitat modification determination for the CRLF based on LOC exceedences. A summary of the risk estimation results are provided in Table 31 for direct and indirect effects to the CRLF and in Table 32 for the PCEs of designated critical habitat for the CRLF.

Table 31. Risk Estimation Summary for Ziram - Direct and Indirect Effects to CRLF.

Assessment Endpoint	LOC Exceedances (Y/N)	Description of Results of Risk Estimation
Aquatic Phase (eggs, larvae, tadpoles, juveniles, and adults)		

Direct Effects Survival, growth, and reproduction of CRLF individuals via direct effects on aquatic phases	Y	LOC exceedences for all use scenarios (almonds, apples/pears, apricots, grapes, peaches and pecans) for acute effects resulted in "may affect" determination. The "No Effect" determination for chronic effects of Thiram to the CRLF is based on no LOC exceedence for any uses.
Indirect Effects Survival, growth, and reproduction of CRLF individuals via effects to food supply (*i.e.*, freshwater invertebrates, non-vascular plants)	Y	LOC exceedences for all use scenarios crops (almonds, apples/pears, apricots, grapes, peaches and pecans) for acute effects resulted in "may affect" determination. The "No Effect" determination for chronic effects of Thiram to the CRLF is based on no LOC exceedence for any uses.
Indirect Effects Survival, growth, and reproduction of CRLF individuals via effects on habitat, cover, and/or primary productivity (*i.e.*, aquatic plant community)	N	There were no LOC exceedences for any use scenarios (almonds, apples/pears, apricots, grapes, peaches and pecans) for vascular or non-vascular aquatic plants. There is an NE determination based on no LOC exceedences.
Indirect Effects Survival, growth, and reproduction of CRLF individuals via effects to riparian vegetation, required to maintain acceptable water quality and habitat in ponds and streams comprising the species' current range.	N	There were no LOC exceedences for any use scenarios (almonds, apples/pears, apricots, grapes, peaches and pecans) for seedling emergence or vegetative vigor studies in terrestrial plants. There is an NE determination based on no LOC exceedences.
Terrestrial Phase (Juveniles and adults)		
Direct Effects Survival, growth, and reproduction of CRLF individuals via direct effects on terrestrial phase adults and juveniles	Y	Due to LOC exceedences for all use scenarios (almonds, apples/pears, apricots, grapes, peaches and pecans) from the T-REX model there is a "may affect" determination for acute direct effects. Due to LOC exceedences for all use scenarios (almonds, apples/pears, apricots, grapes, peaches and pecans) from the T-REX model there is a "may affect" determination for chronic direct effects.
Indirect Effects Survival, growth, and reproduction of CRLF individuals via effects on prey (*i.e.*, terrestrial invertebrates, small terrestrial mammals and terrestrial phase amphibians)	Y	There is potential for indirect prey effects of Ziram on the CRLF consuming terrestrial invertebrates based on potential LOC exceedence. Due to LOC exceedences for use scenarios (almonds, apples/pears, apricots, grapes, peaches and pecans) from the T-REX model calculating RQs for mammals there is a "may affect" determination for acute effects on the CRLF. Due to LOC exceedences for use scenarios (almonds,

		apples/pears, apricots, grapes, peaches and pecans) from the T-REX model calculating RQs for terrestrial-phase amphibians using the bird as a surrogate, there is a "may affect" determination for acute effects on the CRLF.
Indirect Effects Survival, growth, and reproduction of CRLF individuals via effects on habitat (*i.e.*, riparian vegetation)	N	There was no effect at the 25% level for the highest concentration (6 lb/A) tested for terrestrial plants from either seedling emergence or vegetative vigor toxicity tests. Therefore, there is a NE determination.

Table 32. Risk Estimation Summary for Ziram – PCEs of Designated Critical Habitat for the CRLF.

Assessment Endpoint	LOC Exceedances (Y/N)	Description of Results of Risk Estimation
Aquatic Phase PCEs (Aquatic Breeding Habitat and Aquatic Non-Breeding Habitat)		
Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.	N	There was no effect at the 25% level for the highest concentration (6 lb/A) tested for terrestrial plants
Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	N	There were no LOC exceedences for non-vascular or vascular plants. There was no effect at the 25% level for the highest concentration (6 lb/A) tested for terrestrial plants
Alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source.	Y	Based on the LOC exceedences from T-REX for all use scenarios, there is a "habitat modification" determination.
Reduction and/or modification of aquatic-based food sources for pre-metamorphs (e.g., algae)	Y	Based on the LOC exceedences for T-REX for all use scenarios, there is a "habitat modification" determination.
Terrestrial Phase PCEs (Upland Habitat and Dispersal Habitat)		
Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or dripline surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance.	N	There was no effect at the 25% level for the highest concentration (6 lb/A) tested for terrestrial plants
Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat	N	There was no effect at the 25% level for the highest

Assessment Endpoint	LOC Exceedances (Y/N)	Description of Results of Risk Estimation
within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal		concentration (6 lb/A) tested for terrestrial plants
Reduction and/or modification of food sources for terrestrial phase juveniles and adults	y	Based on the LOC exceedences for T-REX for all use scenarios (almonds, apples/pears, apricots, cherries, grapes, peaches and pecans) there is a "habitat modification" determination.
Alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source.	Y	Based on the LOC exceedences from T-REX for all use scenarios (almonds, apples/pears, apricots, cherries, grapes, peaches and pecans), there is a "habitat modification" determination .

Following a "may affect" determination, additional information is considered to refine the potential for exposure at the predicted levels based on the life history characteristics (*i.e.*, habitat range, feeding preferences, etc.) of the CRLF. Based on the best available information, the Agency uses the refined evaluation to distinguish those actions that "may affect, but are not likely to adversely affect" from those actions that are "likely to adversely affect" the CRLF and its designated critical habitat.

The criteria used to make determinations that the effects of an action are "not likely to adversely affect" the CRLF and its designated critical habitat include the following:

- <u>Significance of Effect</u>: Insignificant effects are those that cannot be meaningfully measured, detected, or evaluated in the context of a level of effect where "take" occurs for even a single individual. "Take" in this context means to harass or harm, defined as the following:
    - Harm includes significant habitat modification or degradation that results in death or injury to listed species by significantly impairing behavioral patterns such as breeding, feeding, or sheltering.
    - Harass is defined as actions that create the likelihood of injury to listed species to such an extent as to significantly disrupt normal behavior patterns which include, but are not limited to, breeding, feeding, or sheltering.
- <u>Likelihood of the Effect Occurring</u>: Discountable effects are those that are extremely unlikely to occur.
- <u>Adverse Nature of Effect</u>: Effects that are wholly beneficial without any adverse effects are not considered adverse.

A description of the risk and effects determination for each of the established assessment endpoints for the CRLF and its designated critical habitat is provided in **Sections 5.2.1 through 5.2.3**.

5.2.1 Direct Effects

5.2.1.1 Aquatic-Phase CRLF

The aquatic-phase considers life stages of the frog that are obligatory aquatic organisms, including eggs and larvae. It also considers submerged terrestrial-phase juveniles and adults, which spend a portion of their time in water bodies that may receive runoff and spray drift containing Ziram.

RQs estimated for the direct effect of Ziram on the aquatic-phase CRLF using the fish as a surrogate for all modeled uses exceed the LOC =0.05 for aquatic animals. RQs for acute effects ranged from 1.02 for cherries to 4.5 for wine grapes using a ground application.

The probit analysis was used to refine the acute effects determination. The analysis for probability of an individual effect to the aquatic-phase CRLF based on the slope of the dose response curve for the acute endpoint used to derive the RQ provides additional lines of evidence. The most sensitive toxicity value for fish is for the bluegill sunfish, with an LC50=9.7 μg/L and a slope=6.15.
The chronic effect of Ziram was calculated using the Acute-to-Chronic ratio for Thiram value due to the absence of available chronic Thiram data. There were no chronic RQs for Thiram exceeding the LOC=1 for any use scenarios.

Additional lines of evidence included reviewing open literature for more sensitive survival, growth and reproductive endpoints and reviewing aquatic incident reports from the EIIS database. No open literature provides toxicity values lower than those provided in registrant submitted studies for fish. No aquatic incidents were reported for Ziram in the EIIS database.

Based on no LOC exceedence for chronic effects of Ziram to the CRLF using the fish a as a surrogate, there is a “no effect” determination for direct chronic effects of Ziram on the CRLF.

Direct effects of Ziram on the aquatic-phase CRLF are based on both acute and chronic effects. The “LAA” determination for the aquatic-phase CRLF is based on the “LAA” determination for acute effects.

5.2.1.2 Terrestrial-Phase CRLF

Exposure and risk to the terrestrial phase CRLF is summarized in Table 33. RQs estimated for the direct effect of Ziram on the terrestrial-phase CRLF using the bird as a surrogate for all modeled uses exceed the LOC =0.1 for terrestrial animals.
Acute T-REX RQs for all modeled uses exceed the LOC=0.1. RQs ranged from 30.66 for cherries to 86.93 for peaches.

Chronic T-REX RQs for all modeled uses exceed the LOC =1 using the bird as a surrogate for the CRLF. RQs range from 64.87 for cherries to 183.92 for peaches.

Results of T-HERPS for Direct Effects Using the Bird as a Surrogate

T-HERPS is used to refine the results from the T-REX model. RQs are based on results using the bird consuming small insects as a surrogate for direct effects on the CRLF. Examples of T-HERPS calculations are found in Appendix K.

Table 33. Summary of Direct Effect T-HERP RQs for the Terrestrial-phase CRLF.

Use Scenario	Surrogate Species	Toxicity Value (mg/L)	Dose-based EEC (mg/L)[b]	Dose-based RQ	Dietary-based EEC (mg/L)[b]	Dietary-based RQ
Acute Direct Toxicity LD50						
Almonds	Bird-quail	97	117.32	**1.21**	3020	**0.59**
Apples/Pears			88.47	**0.91**	2277	**0.44**
Apricots			117.32	**1.21**	3020	**0.59**
Cherries			73.09	**0.75**	1881	**0.36**
All Grapes			75.49	**0.78**	1943	**0.38**
Peaches			207.22	**2.14**	5334	**1.03**
Pecans			86.28	**0.89**	2221	**0.43**
Chronic Direct Toxicity NOAEC						
Almonds	Bird-mallard	9.6			3020	**104.13**
Apples/Pears					2277	**78.52**
Apricots					3020	**104.13**
Cherries					1881	**64.87**
All Grapes					1943	**67.00**
Peaches					5334	**183.92**
Pecans					2221	**76.58**

[a]RQs associated with acute and chronic direct toxicity to the CRLF are also used to assess potential indirect effects to the CRLF based on a reduction in freshwater fish and frogs as food items.
[b]The highest EEC based on Ziram use on peaches at 7.6 lb ai/A/acre/year.
[c]The probit slope value for the acute quail toxicity test is the 4.5 default slope.
Bold: Exceeds Acute endangered species LOC of 0.1 and the chronic LOC=1.

Both dose-based and dietary RQs were calculated to assess the direct effects of Ziram on the CRLF using the bird as a surrogate. There were LOC exceedences for all use scenarios for acute dose-based RQs for direct effects. RQs ranged from 0.75 for cherries to 2.14 for peaches. In addition to the dose-based RQs, dietary-base RQS also exceeded the LOC=0.1 for all uses.

Direct effects to the CRLF also include chronic effects. There were LOC exceedences for all use scenarios for chronic dietary-based RQs for direct effects. RQs ranged from 64.87 for cherries to 183.92 for peaches.

The probit model was used to refine the results for acute effects. The analysis for probability of an individual effect to the terrestrial-phase CRLF based on the slope of the dose response curve for the acute endpoint used to derive the RQ provides additional lines of evidence. The most sensitive toxicity value for quail is an LD50=97 mg/kg with a default slope=4.5. The chance of an individual effect =1.00 for all modeled uses.

There were no open literature toxicity values more sensitive than those in the registrant submitted studies for Ziram. There were no bird incidents reported in the EIIS database for Ziram. However there was one bird incident (I005754-012) reported for Thiram. The report from Bakersfield, California of 200 dead cedar waxwings was classified as unlikely based on no pesticide residue detected in samples.

Results form the T-HERPs and probit models, open literature data, as well as the incident reports provide the basis for lines of evidence in the effects determination. Based on the lines-of-evidence, there is a potential direct impact to the terrestrial-phase of the CRLF based on this endpoint.

5.2.2 Indirect Effects (Reductions in Prey Base)

5.2.2.1 Algae (non-vascular plants)

As discussed in Section 2.5.3, the diet of CRLF tadpoles is composed primarily of unicellular aquatic plants (i.e., algae and diatoms) and detritus. RQs for non-vascular aquatic plants range from 0.15 for cherries to 0.65 for wine grapes with a ground application. All use scenarios resulted in acute RQs < 1, therefore Ziram is not likely to indirectly affect the CRLF. There is no NAWQA monitoring data available for Ziram. There is no open literature with more sensitive results than registered submitted studies. Based on the lines-of-evidence there is not a potential indirect impact to the CRLF based on this endpoint.

5.2.2.2 Aquatic Invertebrates

The potential for Ziram to elicit indirect effects to the CRLF via effects on freshwater invertebrate food items is dependent on several factors including: (1) the potential magnitude of effect on freshwater invertebrate individuals and populations; and (2) the number of prey species potentially affected relative to the expected number of species needed to maintain the dietary needs of the CRLF. Together, these data provide a basis to evaluate whether the number of individuals within a prey species is likely to be reduced such that it may indirectly affect the CRLF.

All use scenarios resulted in LOC exceedences for acute indirect dietary effects of Ziram on the CRLF. RQs ranged from 0.21 for cherries to 0.91 for peaches

No modeled chronic RQs exceeded the LOC=1. RQs ranged from 0.01 for peaches and pecans to 0.2 for wine grapes.

The percentage effect to the aquatic invertebrates as a prey base from the probit model is based on the slope of the dose response curve (1.99 for the acute endpoint used to derive the RQ. The results for the indirect effect of a CRLF consuming aquatic invertebrates range from -0.08 for wine grapes with a ground application to -1.16 for cherries.

There were no open literature toxicity values more sensitive than the registrant submitted studies. There is no monitoring data from the NAQWA database available for comparison with modeled exposure concentrations. Based on the lines-of-evidence, there is a potential indirect impact to the CRLF based on this endpoint.

5.2.2.3 Fish and Aquatic-phase Frogs

Based on the lines of evidence using the fish as a surrogate for the CRLF, there are potential indirect impacts to the CRLF. The percentage effect to the mammalian prey base is from the probit model based on the slope of the dose response curve (6.15) for the acute endpoint used to derive the RQ.

5.2.2.4 Terrestrial Invertebrates

When the terrestrial-phase CRLF reaches juvenile and adult stages, its diet is mainly composed of terrestrial invertebrates. To determine the risk for prey reduction, RQs were estimated for small and large insects, and then the chance of individual effect was modeled using the IEC program. Due to all modeled uses for both small and large insects resulting in acute upper-bound RQs for Ziram resulting in RQs higher than the LOC=0.05, there is a “may affect” determination. The probit analysis was not used for Ziram due to the indeterminate LD50. Additional support for the “LAA” determination is based on the Thiram analysis using the more sensitive LD50=73.72. Thiram RQs for small insects range from 3.27 for cherries to 9.26 for peaches resulting in support for the “LAA” determination. RQs for large insects range from 0.36 for cherries to 1.03 for peaches.

The probit was used to refine the analysis for terrestrial insects. There was a significant effect based on the individual probability for small insects for Thiram. RQs for all uses for small insects exceeded the acute risk LOC=0.5. The chance of individual effect ranges from about 1 in 1.15 for nectarines/peaches to about 1 in 1.37 for cherries.

There is also a significant chance of individual effects for large insects for Thiram. The chance of individual effect ranges from about 1 in 1.98for peaches to about 1 in 3.33 for cherries.

Additional information is provided from open literature review and a review of incident reports from the EIIS database. No additional toxicity values were available from open literature. No terrestrial invertebrate incidents were reported for Ziram or Thiram.

Therefore, there is a LAA: determination for the CRLF consuming terrestrial invertebrates based on RQs exceeding the LOC for small insects for all uses. Supporting lines of evidence for large insects are based on the Thiram analysis as well as no reported terrestrial invertebrate incidents from the EIIS database.

5.2.2.4 Mammals

Life history data for terrestrial-phase CRLFs indicate that large adult frogs consume terrestrial vertebrates, including mice. RQs for indirect effects for the CRLF consuming small mammals range from 4.53 for cherries to 12.85 for peaches. Based on acute RQs > 0.5 for all use scenarios, Ziram results in an “LAA” determination.

Supporting evidence is provided by the probit analysis. All modeled uses resulted in significant chance of reduction in prey population based on the probit analysis using the default slope=4.5. All uses resulted in a 1 in 1.00 chance of an individual effect.

There are no open literature toxicity values more sensitive than registrant submitted data. There are also no mammal incidents reported in the EIIS database for either Ziram or Thiram.

Lines of evidence such as open literature and incident reports provide additional information for the determination. The lines-of-evidence support the “LAA” determination.

5.2.2.5 Terrestrial-phase Amphibians

Terrestrial-phase adult CRLFs also consume frogs. Avian RQ values representing direct exposures of Ziram to terrestrial-phase CRLFs are used to represent exposures of Ziram to frogs in terrestrial habitats.

There were LOC exceedences for all use scenarios for acute dose-based RQs for direct effects. RQs ranged from 0.75 for cherries to 2.14 for nectarines/peaches. Based on RQs exceeding the acute risk LOC=0.5, there is an “LAA” determination.

The probit results support the “LAA” determination. Based on the probit analysis there was a 1 in 1.00 chance of individual effect for all use scenarios.

There were LOC exceedences for all use scenarios for chronic dietary-based RQs for direct effects. RQs ranged from 64.87 for cherries to 183.92 for peaches.

5.2.3 Indirect Effects (via Habitat Effects)

5.2.3.1 Aquatic Plants (Vascular and Non-vascular)

Aquatic plants serve several important functions in aquatic ecosystems. Non-vascular aquatic plants are primary producers and provide the autochthonous energy base for aquatic ecosystems. Vascular plants provide structure as attachment sites and refugia for many aquatic invertebrates, fish, and juvenile organisms, such as fish and frogs. In addition, vascular plants also provide primary productivity and oxygen to the aquatic ecosystem. Rooted plants help reduce sediment loading and provide stability to nearshore areas and lower streambanks. In addition, vascular aquatic plants are important as attachment sites for egg masses of CRLFs.

Potential indirect effects to the CRLF based on impacts to habitat and/or primary production were assessed using RQs from freshwater aquatic vascular and non-vascular plant data. No RQs exceeded the LOC for any use scenarios for aquatic vascular plants.

In addition to aquatic vascular plants, an analysis of the effect of Ziram on non-vascular aquatic plants was conducted.

No RQs exceeded the LOC for any use scenarios for aquatic non-vascular plants. No surface water monitoring data is available for Ziram. There was a “no effect” determination for the indirect effects of aquatic-plants based on no LOC exceedence.

There is no open literature with a more sensitive endpoint than the registrant submitted studies. There are no reported aquatic plant incidents in the EIIS database.

5.2.3.2 Terrestrial Plants

Terrestrial plants serve several important habitat-related functions for the CRLF. In addition to providing habitat and cover for invertebrate and vertebrate prey items of the CRLF, terrestrial vegetation also provides shelter for the CRLF and cover from predators while foraging. Terrestrial plants also provide energy to the terrestrial ecosystem through primary production. Upland vegetation including grassland and woodlands provides cover during dispersal. Riparian vegetation helps to maintain the integrity of aquatic systems by providing bank and thermal stability, serving as a buffer to filter out sediment, nutrients, and contaminants before they reach the watershed, and serving as an energy source.

Results from terrestrial plant studies for seedling emergence and vegetative vigor were evaluated. A Tier 1 seedling emergence toxicity tests for terrestrial plants indicated no effect at the 25% threshold for any use scenarios for either monocots or dicots. A Tier 1 vegetative vigor study resulted in no effects at the 25% threshold at the highest concentration tested for either monocots or dicots.
There are no open literature endpoints more sensitive than those in registrant submitted studies. There are no reported terrestrial plant incidents for Ziram in the EIIS database.
Based on the lines-of-evidence, there is not a potential indirect impact to the CRLF based on this endpoint.

5.2.4 Modification to Designated Critical Habitat

5.2.4.1 Aquatic-Phase PCEs

Three of the four assessment endpoints for the aquatic-phase primary constituent elements (PCEs) of designated critical habitat for the CRLF are related to potential effects to aquatic and/or terrestrial plants:

- Alteration of channel/pond morphology or geometry and/or increase in sediment deposition within the stream channel or pond: aquatic habitat (including riparian vegetation) provides for shelter, foraging, predator avoidance, and aquatic dispersal for juvenile and adult CRLFs.

- Alteration in water chemistry/quality including temperature, turbidity, and oxygen content necessary for normal growth and viability of juvenile and adult CRLFs and their food source.
- Reduction and/or modification of aquatic-based food sources for pre-metamorphs (*e.g.,* algae).

Conclusions for potential indirect effects to the CRLF via direct effects to aquatic and terrestrial plants are used to determine whether modification to critical habitat may occur. There is not a potential for habitat modification via impacts to aquatic plants (Sections 5.2.2.1 and 5.2.3.1) or terrestrial plants (5.2.3.2)

The remaining aquatic-phase PCE is "alteration of other chemical characteristics necessary for normal growth and viability of CRLFs and their food source." Other than impacts to algae as food items for tadpoles (discussed above), this PCE is assessed by considering direct and indirect effects to the aquatic-phase CRLF via acute and chronic freshwater fish and invertebrate toxicity endpoints as measures of effects. There is a potential for habitat modification via impacts to aquatic-phase CRLFs (Sections 5.2.1.1) and effects to freshwater invertebrates and fish as food items (Sections 5.2.2.2 and 5.2.2.3).

5.2.4.2 Terrestrial-Phase PCEs

Two of the four assessment endpoints for the terrestrial-phase PCEs of designated critical habitat for the CRLF are related to potential effects to terrestrial plants:

- Elimination and/or disturbance of upland habitat; ability of habitat to support food source of CRLFs: Upland areas within 200 ft of the edge of the riparian vegetation or drip line surrounding aquatic and riparian habitat that are comprised of grasslands, woodlands, and/or wetland/riparian plant species that provides the CRLF shelter, forage, and predator avoidance.

- Elimination and/or disturbance of dispersal habitat: Upland or riparian dispersal habitat within designated units and between occupied locations within 0.7 mi of each other that allow for movement between sites including both natural and altered sites which do not contain barriers to dispersal.

There is not a potential for habitat modification via impacts to terrestrial plants (5.2.3.2).

The third terrestrial-phase PCE is "reduction and/or modification of food sources for terrestrial phase juveniles and adults." To assess the impact of Ziram on this PCE, acute and chronic toxicity endpoints for terrestrial invertebrates, mammals, and terrestrial-phase frogs are used as measures of effects. There is a potential for habitat modification via indirect effects to terrestrial-phase CRLFs via reduction in prey base (Section 5.2.2.4 for terrestrial invertebrates, Section 5.2.2.5 for mammals, and 5.2.2.6 for frogs).

The fourth terrestrial-phase PCE is based on alteration of chemical characteristics necessary for normal growth and viability of juvenile and adult CRLFs and their food source. There is a potential for habitat modification via direct (Section 5.2.1.2) and indirect effects (Sections 5.2.2.4, 5.2.2.5, and 5.2.2.6) to terrestrial-phase CRLFs.

5.3 Action Area Spatial Analysis

Appendix L provides an overview of where the action area overlaps with species range as described in Section 2.5.1. The overlap is illustrated in Figure 10. The analysis indicates that overlap between the Ziram action area and species range (defined by critical habitat, core areas, and CNDDB occurrence data) occurs in all eight of the CRLF Recovery Units.

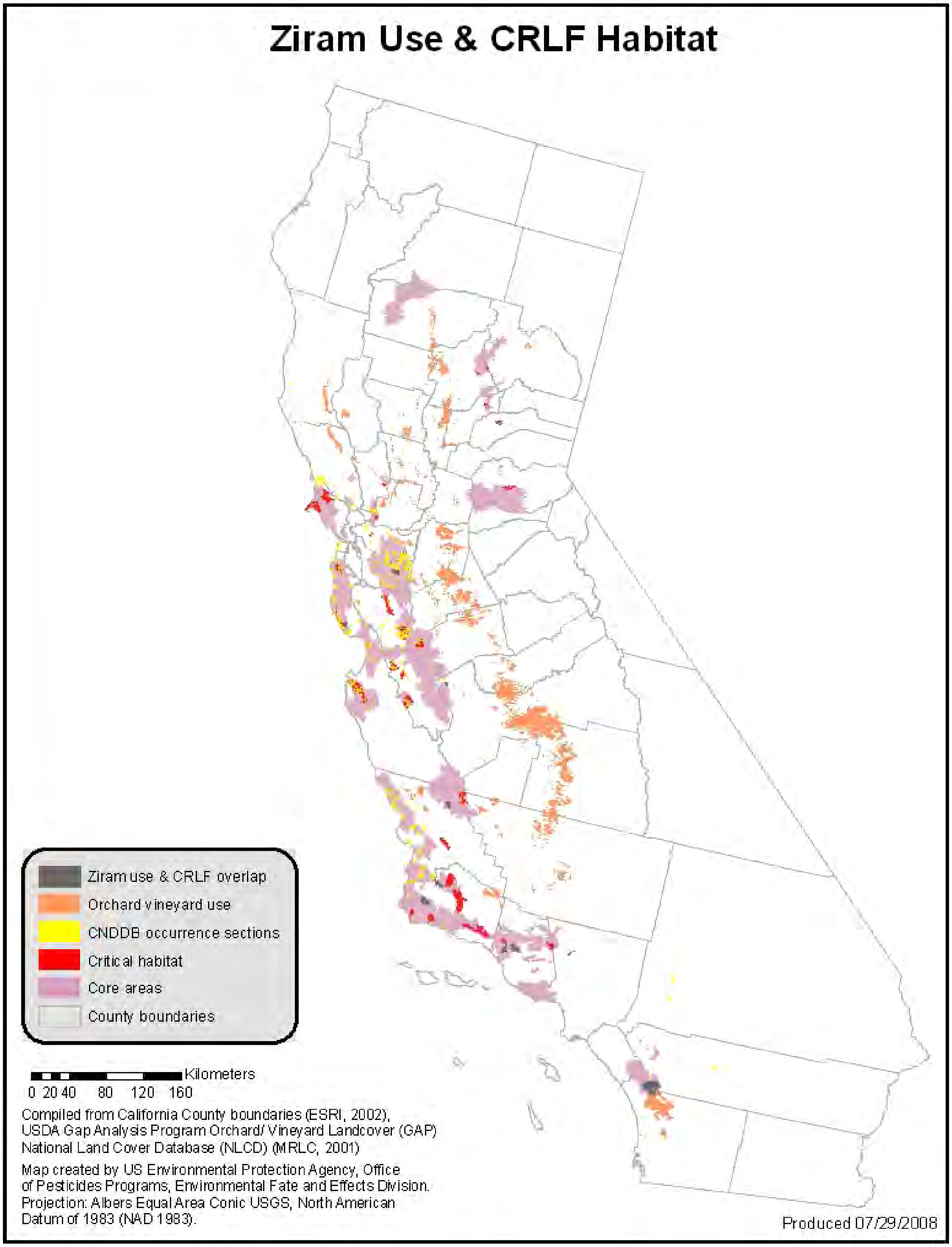


Figure 10. Ziram Use and CRLF habitat overlap.

Pesticide exposures and predicted risks to the species and its resources (i.e., food and habitat) are not expected to be uniform across the action area. In fact, given the assumptions of drift and downstream transport (i.e., attenuation with distance), pesticide exposure and associated risks to the species and its resources are expected to decrease with increasing distance away from the treated field or site of application. That is, areas where overlap occurs between the initial area of concern and the species range are where the risk is presumed to be greatest. Moving from the initial area of concern to the edge of the action area, whether it be defined by spray drift distances or by transport of Ziram downstream from the site of application, the magnitude of exposure decreases as does the potential risk. For example, the action area is defined as the entire state of California since Ziram is identified as a carcinogen (Section 2.7).

6. Uncertainties

6.1 Exposure Assessment Uncertainties

6.1.1 Maximum Use Scenario

The screening-level risk assessment focuses on characterizing potential ecological risks resulting from a maximum use scenario, which is determined from labeled statements of maximum application rate and number of applications with the shortest time interval between applications. The frequency at which actual uses approach this maximum use scenario may be dependant on pest resistance, timing of applications, cultural practices, and market forces.

6.1.2 Aquatic Exposure Modeling of Ziram

The standard ecological water body scenario (EXAMS pond) used to calculate potential aquatic exposure to pesticides is intended to represent conservative estimates, and to avoid underestimations of the actual exposure. The standard scenario consists of application to a 10-hectare field bordering a 1-hectare, 2-meter deep (20,000 m^3) pond with no outlet. Exposure estimates generated using the EXAMS pond are intended to represent a wide variety of vulnerable water bodies that occur at the top of watersheds including prairie pot holes, playa lakes, wetlands, vernal pools, man-made and natural ponds, and intermittent and lower order streams. As a group, there are factors that make these water bodies more or less vulnerable than the EXAMS pond. Static water bodies that have larger ratios of pesticide-treated drainage area to water body volume would be expected to have higher peak EECs than the EXAMS pond. These water bodies will be either smaller in size or have larger drainage areas. Smaller water bodies have limited storage capacity and thus may overflow and carry pesticide in the discharge, whereas the EXAMS pond has no discharge. As watershed size increases beyond 10-hectares, it becomes increasingly unlikely that the entire watershed is planted with a single crop that is all treated simultaneously with the pesticide. Headwater streams can also have peak concentrations higher than the EXAMS pond, but they likely persist for only short periods of time and are then carried and dissipated downstream.

The Agency acknowledges that there are some unique aquatic habitats that are not accurately captured by this modeling scenario and modeling results may, therefore, under- or over-estimate exposure, depending on a number of variables. For example, aquatic-phase CRLFs may inhabit water bodies of different size and depth and/or are located adjacent to larger or smaller drainage areas than the EXAMS pond. The Agency does not currently have sufficient information regarding the hydrology of these aquatic habitats to develop a specific alternate scenario for the CRLF. CRLFs prefer habitat with perennial (present year-round) or near-perennial water and do not frequently inhabit vernal (temporary) pools because conditions in these habitats are generally not suitable (Hayes and Jennings 1988). Therefore, the EXAMS pond is assumed to be representative of exposure to aquatic-phase CRLFs. In addition, the Services agree that the existing EXAMS pond represents the best currently available approach for estimating aquatic exposure to pesticides (USFWS/NMFS 2004).

In general, the linked PRZM/EXAMS model produces estimated aquatic concentrations that are expected to be exceeded once within a ten-year period. The Pesticide Root Zone Model is a process or "simulation" model that calculates what happens to a pesticide in an agricultural field on a day-to-day basis. It considers factors such as rainfall and plant transpiration of water, as well as how and when the pesticide is applied. It has two major components: hydrology and chemical transport. Water movement is simulated by the use of generalized soil parameters, including field capacity, wilting point, and saturation water content. The chemical transport component can simulate pesticide application on the soil or on the plant foliage. Dissolved, adsorbed, and vapor-phase concentrations in the soil are estimated by simultaneously considering the processes of pesticide uptake by plants, surface runoff, erosion, decay, volatilization, foliar wash-off, advection, dispersion, and retardation.

Uncertainties associated with each of these individual components add to the overall uncertainty of the modeled concentrations. Additionally, model inputs from the environmental fate degradation studies are chosen to represent the upper confidence bound on the mean values that are not expected to be exceeded in the environment approximately 90 percent of the time. Mobility input values are chosen to be representative of conditions in the environment. The natural variation in soils adds to the uncertainty of modeled values. Factors such as application date, crop emergence date, and canopy cover can also affect estimated concentrations, adding to the uncertainty of modeled values. Factors within the ambient environment such as soil temperatures, sunlight intensity, antecedent soil moisture, and surface water temperatures can cause actual aquatic concentrations to differ for the modeled values.

Unlike spray drift, tools are currently not available to evaluate the effectiveness of a vegetative setback on runoff and loadings. The effectiveness of vegetative setbacks is highly dependent on the condition of the vegetative strip. For example, a well-established, healthy vegetative setback can be a very effective means of reducing runoff and erosion from agricultural fields. Alternatively, a setback of poor vegetative quality or a setback that is channelized can be ineffective at reducing loadings. Until such time as a quantitative method to estimate the effect of vegetative setbacks on various conditions on pesticide loadings becomes available, the aquatic exposure predictions are likely to overestimate exposure where healthy vegetative setbacks exist and underestimate exposure where poorly developed, channelized, or bare setbacks exist.

6.1.3 Action Area Uncertainties

An example of an important simplifying assumption that may require future refinement is the assumption of uniform runoff characteristics throughout a landscape. It is well documented that runoff characteristics are highly non-uniform and anisotropic, and become increasingly so as the area under consideration becomes larger. The assumption made for estimating the aquatic action area (based on predicted in-stream dilution) was that the entire landscape exhibited runoff properties identical to those commonly found in agricultural lands in this region. However, considering the vastly different runoff characteristics of: a) undeveloped (especially forested) areas, which exhibit the least amount of surface runoff but the greatest amount of groundwater recharge; b) suburban/residential areas, which are dominated by the relationship between impermeable surfaces (roads, lots) and grassed/other areas (lawns) plus local drainage management; c) urban areas, that are dominated by managed storm drainage and impermeable surfaces; and d) agricultural areas dominated by Hortonian and focused runoff (especially with row crops), a refined assessment should incorporate these differences for modeled stream flow generation. As the zone around the immediate (application) target area expands, there will be greater variability in the landscape; in the context of a risk assessment, the runoff potential that is assumed for the expanding area will be a crucial variable (since dilution at the outflow point is determined by the size of the expanding area). Thus, it important to know at least some approximate estimate of types of land use within that region. Runoff from forested areas ranges from 45 – 2,700% less than from agricultural areas; in most studies, runoff was 2.5 to 7 times higher in agricultural areas (e.g., Okisaka et al., 1997; Karvonen et al., 1999; McDonald et al., 2002; Phuong and van Dam 2002). Differences in runoff potential between urban/suburban areas and agricultural areas are generally less than between agricultural and forested areas. In terms of likely runoff potential (other variables – such as topography and rainfall – being equal), the relationship is generally as follows (going from lowest to highest runoff potential):

Three-tiered forest < agroforestry < suburban < row-crop agriculture < urban.

There are, however, other uncertainties that should serve to counteract the effects of the aforementioned issue. For example, the dilution model considers that 100% of the agricultural area has the chemical applied, which is almost certainly a gross over-estimation. Thus, there will be assumed chemical contributions from agricultural areas that will actually be contributing only runoff water (dilutant); so some contributions to total contaminant load will really serve to lessen rather than increase aquatic concentrations. In light of these (and other) confounding factors, Agency believes that this model gives us the best available estimates under current circumstances.

6.1.4 Usage Uncertainties

County-level usage data were obtained from California's Department of Pesticide Regulation Pesticide Use Reporting (CDPR PUR) database. Four years of data (2002 – 2005) were included in this analysis because statistical methodology for identifying outliers, in terms of area treated and pounds applied, was provided by CDPR for these years only. No methodology for removing outliers was provided by CDPR for 2001 and earlier pesticide data; therefore, this information was not included in the analysis because it may misrepresent actual usage patterns. CDPR PUR documentation indicates that errors in the data may include the following: a misplaced decimal; incorrect measures, area treated, or units; and reports of diluted pesticide concentrations. In addition, it is possible that the data may contain reports for pesticide uses that have been cancelled. The CPDR PUR data does not include home-owner applied pesticides; therefore, residential uses are not likely to be reported. As with all pesticide usage data, there may be instances of misuse and misreporting. The Agency made use of the most current, verifiable information; in cases where there were discrepancies, the most conservative information was used.

6.1.5 Terrestrial Exposure Modeling of Ziram

The Agency relies on the work of Fletcher et al. (1994) for setting the assumed pesticide residues in wildlife dietary items. These residue assumptions are believed to reflect a realistic upper-bound residue estimate, although the degree to which this assumption reflects a specific percentile estimate is difficult to quantify. It is important to note that the field measurement efforts used to develop the Fletcher estimates of exposure involve highly varied sampling techniques. It is entirely possible that much of these data reflect residues averaged over entire above ground plants in the case of grass and forage sampling.

It was assumed that ingestion of food items in the field occurs at rates commensurate with those in the laboratory. Although the screening assessment process adjusts dry-weight estimates of food intake to reflect the increased mass in fresh-weight wildlife food intake estimates, it does not allow for gross energy differences. Direct comparison of a laboratory dietary concentration-based effects threshold to a fresh-weight pesticide residue estimate would result in an underestimation of field exposure by food consumption by a factor of 1.25 – 2.5 for most food items.

Differences in assimilative efficiency between laboratory and wild diets suggest that current screening assessment methods do not account for a potentially important aspect of food requirements. Depending upon species and dietary matrix, bird assimilation of wild diet energy ranges from 23 – 80%, and mammal's assimilation ranges from 41 – 85% (U.S. Environmental Protection Agency, 1993). If it is assumed that laboratory chow is formulated to maximize assimilative efficiency (e.g., a value of 85%), a potential for underestimation of exposure may exist by assuming that consumption of food in the wild is comparable with consumption during laboratory testing. In the screening process, exposure may be underestimated because metabolic rates are not related to food consumption.

For the terrestrial exposure analysis of this risk assessment, a generic bird or mammal was assumed to occupy either the treated field or adjacent areas receiving a treatment rate on the field. Actual habitat requirements of any particular terrestrial species were not considered, and it was assumed that species occupy, exclusively and permanently, the modeled treatment area. Spray drift model predictions suggest that this assumption leads to an overestimation of exposure to species that do not occupy the treated field exclusively and permanently.

6.1.6 Spray Drift Modeling

Although there may be multiple Ziram applications at a single site, it is unlikely that the same organism would be exposed to the maximum amount of spray drift from every application made.

In order for an organism to receive the maximum concentration of Ziram from multiple applications, each application of Ziram would have to occur under identical atmospheric conditions (e.g., same wind speed and same wind direction) and (if it is an animal) the animal being exposed would have to be present in the same location (which receives the maximum amount of spray drift) after each application. Although there may be sites where the dominant wind direction is fairly consistent (at least during the relatively quiescent conditions that are most favorable for aerial spray applications), it is nevertheless highly unlikely that any specific area would receive the maximum amount of spray drift repeatedly. It appears that in most areas (based upon available meteorological data) wind direction is temporally very changeable, even within the same day. Additionally, other factors, including variations in topography, cover, and meteorological conditions over the transport distance are not accounted for by the AgDRIFT/AGDISP model (*i.e*., it models spray drift from aerial and ground applications in a flat area with little to no ground cover and a steady, constant wind speed and direction). Therefore, in most cases, the drift estimates from AgDRIFT/AGDISP may overestimate exposure even from single applications, especially as the distance increases from the site of application, since the model does not account for potential obstructions (*e.g*., large hills, berms, buildings, trees, *etc*.). Furthermore, conservative assumptions are often made regarding the droplet size distributions being modeled ('ASAE Very Fine to Fine' for orchard uses and 'ASAE Very Fine' for agricultural uses), the application method (*i.e*., aerial), release heights and wind speeds. Alterations in any of these inputs would change the area of potential effect.

6.2 Effects Assessment Uncertainties

6.2.1 Age Class and Sensitivity of Effects Thresholds

It is generally recognized that test organism age may have a significant impact on the observed sensitivity to a toxicant. The acute toxicity data for fish are collected on juvenile fish between 0.1 and 5 grams. Aquatic invertebrate acute testing is performed on recommended immature age classes (e.g., first instar for daphnids, second instar for amphipods, stoneflies, mayflies, and third instar for midges).

Testing of juveniles may overestimate toxicity at older age classes for pesticide active ingredients that act directly without metabolic transformation because younger age classes may not have the enzymatic systems associated with detoxifying xenobiotics. In so far as the available toxicity data may provide ranges of sensitivity information with respect to age class, this assessment uses the most sensitive life-stage information as measures of effect for surrogate aquatic animals, and is therefore, considered as protective of the CRLF.

6.2.2 Use of Surrogate Species Effects Data

Guideline toxicity tests and open literature data on Ziram are not available for frogs or any other aquatic-phase amphibian; therefore, freshwater fish are used as surrogate species for aquatic-phase amphibians. Therefore, endpoints based on freshwater fish ecotoxicity data are assumed to be protective of potential direct effects to aquatic-phase amphibians including the CRLF, and extrapolation of the risk conclusions from the most sensitive tested species to the aquatic-phase CRLF is likely to overestimate the potential risks to those species. Efforts are made to select the organisms most likely to be affected by the type of compound and usage pattern; however, there is an inherent uncertainty in extrapolating across phyla. In addition, the Agency's LOCs are intentionally set very low, and conservative estimates are made in the screening level risk assessment to account for these uncertainties.

6.2.3 Sublethal Effects

When assessing acute risk, the screening risk assessment relies on the acute mortality endpoint as well as a suite of sublethal responses to the pesticide, as determined by the testing of species response to chronic exposure conditions and subsequent chronic risk assessment. Consideration of additional sublethal data in the effects determination t is exercised on a case-by-case basis and only after careful consideration of the nature of the sublethal effect measured and the extent and quality of available data to support establishing a plausible relationship between the measure of effect (sublethal endpoint) and the assessment endpoints. However, the full suite of sublethal effects from valid open literature studies is considered for the purposes of defining the action area.

To the extent to which sublethal effects are not considered in this assessment, the potential direct and indirect effects of Ziram on CRLF may be underestimated.

6.2.4 Location of Wildlife Species

For the terrestrial exposure analysis of this risk assessment, a generic bird or mammal was assumed to occupy either the treated field or adjacent areas receiving a treatment rate on the field. Actual habitat requirements of any particular terrestrial species were not considered, and it was assumed that species occupy, exclusively and permanently, the modeled treatment area. Spray drift model predictions suggest that this assumption leads to an overestimation of exposure to species that do not occupy the treated field exclusively and permanently.

7. Risk Conclusions

In fulfilling its obligations under Section 7(a)(2) of the Endangered Species Act, the information presented in this endangered species risk assessment represents the best data currently available to assess the potential risks of Ziram to the CRLF and its designated critical habitat.

Based on the best available information, the Agency makes a May Affect, Likely to Adversely Affect determination for the CRLF from the use of Ziram. Additionally, the Agency has determined that there is the potential for modification of CRLF designated critical habitat from the use of Ziram. All aquatic-phase modeled acute exposures for Ziram resulted in LOCs exceeding the endangered species thresholds for direct effects using the fish as a surrogate for the CRLF. Therefore, there is a "may affect" for Ziram based on those estimates

Using the probit analysis for Ziram as a refinement, the results indicate there is the potential for an individual effect for all use scenarios. Probit analysis indicates a chance of individual effects ranging about 1 in 1 for almonds with an aerial application, apricots with a ground application, apricots with an aerial application, nectarines with a ground application, nectarines with an aerial application, peaches with a ground application, peaches with an aerial application, pecans wine grapes with a ground application and wine grapes with an aerial application to 1.39 for cherries with an aerial application.

In addition to the probit refinement, the EIIS Incident database was reviewed for aquatic incidents. However, no aquatic incidents were reported.

Additional support for the “LAA” determination is based on acute Thiram results. Probit analysis indicates a chance of individual effects ranging about 1 in 1.00 for wine grapes with an aerial application to 4.12 for pears.
All terrestrial-phase modeled acute exposures for Ziram resulted in LOCs exceeding the endangered species thresholds for direct effects using the bird as a surrogate for the CRLF. Therefore, there is a “may affect” for Ziram based on those estimates. An “LAA” determination was based on T-HERPS results, with acute dose-based RQs exceeding the LOC for all use scenarios.

Additional lines of evidence supporting the “LAA” determination include the probit refinement as well as a review of open literature and terrestrial incidents. Using the probit analysis as a refinement, the results indicate there is the potential for an individual effect for all use scenarios supporting to an “LAA” determination. The chance of individual effects for all uses was about 1 in 1.00.

In addition to the probit refinement, a review of open literature resulted in no endpoints that were more sensitive than those in registrant submitted studies. A review of terrestrial incidents in the EIIS database resulted in no reports of terrestrial animal incidents for Ziram and one report of bird mortality for Thiram. However, this Thiram report was classified as unlikely due to no pesticide detected in the birds

The direct effects assessment also included reviewing the chronic effects of Ziram on the CRLF. Due to the persistence of the degradate Thiram, Thiram toxicity values will be used with Ziram exposures for this assessment. All chronic dose-based RQs also exceeded the LOC for all use scenarios for both T-REX and T-HERPS supporting an “LAA” determination.

Indirect acute dietary RQs for the aquatic-phase CRLF also exceeded the Endangered Species LOC=0.05 for all uses for aquatic invertebrates as prey items resulting in a “may affect”. The probit analysis indicates there is a potential chance for individual effect for all uses except cherries (about 1 in 11.3 probit result). The “LAA” determination is based on the probit analysis using the slope=1.99 with significant results ranging from about 1 in 2.14 for wine grapes with a ground application to about 1 in 8.19 for grapes.

The “LAA” determination for indirect acute effects for the CRLF is supported by no aquatic incidents for either Ziram or Thiram reported in the EIIS database. There no Ziram aquatic endpoints more sensitive than the registrant submitted studies available in open literature.

Indirect dietary effects for the terrestrial-phase CRLF exceed the LOC threshold for all use scenarios, including fish/frog, terrestrial invertebrates and small mammals resulting in a "may affect" for modeled species.

Using fish as prey for the CRLF, all modeled acute exposures for Ziram resulted in LOCs exceeding the endangered species thresholds for indirect effects. Therefore, there is a "may affect" for Ziram based on those estimates. An "LAA" determination is based on RQs exceeding the acute risk LOC=0.5 for indirect effects on the CRLF consuming fish for all uses. RQs range from 1.66 for cherries with an aerial application to 5.46 for wine grapes with a ground application. Using the probit analysis as a refinement, the results indicate there is the potential for an individual effect for all use scenarios.

In addition to the probit refinement, a review of the open literature resulted in no endpoints more sensitive than the registrant submitted studies. The EIIS Incident database was reviewed for aquatic incidents with the potential to affect fish as prey items. However, no Ziram aquatic incidents were reported.

In addition to the Ziram analysis, results of Thiram acute analysis also support an "LAA" determination based on acute risk LOC exceedence. RQs range from 4.88 for pears to 41.26 for nectarines.

Using the probit analysis as a refinement, the results indicate there is the potential for an individual effect for all use scenarios. Chance of individual effects range from about 1 in 1 for nectarines, peaches and wine grapes with an aerial application to 4.12 for pears

Other supporting lines of evidence include incident reports. No reported Thiram aquatic animal incidents were reported in the EIIS database.

Indirect dietary effects for the terrestrial-phase CRLF exceed the LOC threshold = 0.05 for all use scenarios for both small and large insects for both Ziram, resulting in a "may affect" determination. There is an "LAA" determination for the effects of Ziram on terrestrial invertebrates consumed by the CRLF based on upper-bound RQs exceeding the LOC=0.5.

Due to the indeterminate endpoint for the Ziram study, additional analysis for lines of evidence included an analysis of the Thiram. The Thiram probit analysis using the default slope=4.5 was used to further refine the determination. The probit analysis indicated significant effects based on the chance of individual effects for all uses. The chance of individual effects ranged from about 1 in to about 1 in 2.56 for almonds and apricots to about 1 in 3.33 for cherries. The results of the Thiram analysis support the "LAA" determination.

A review of ECOTOX resulted in no open literature studies with more sensitive endpoints for Ziram. No terrestrial invertebrate incidents were reported in the EIIS database.

Analysis of indirect dietary effects for the CRLF consuming small mammals resulted in T-REX RQs exceeding the LOC threshold for acute effects. This resulted in a "may affect" determination. The "LAA" determination for indirect effects on the CRLF consuming small mammals is based on acute RQs exceeding the acute risk LOC=0.5 for all uses for Ziram. RQs range from 4.53 for cherries to 12.85 for nectarines/peaches.

A review of the open literature for mammals resulted in no endpoints more sensitive than the registrant submitted studies. The EIIS incident database was reviewed for terrestrial incidents with the potential to affect mammals as prey items. However, no Ziram terrestrial animal incidents were reported.

In addition to acute effects, chronic effects for mammals as prey are also analyzed. RQs exceed the LOC=1 for all uses. Spatial analysis is a refinement for the chronic effect. There is an overlap between areas of the expected adverse affect and where the species is located; therefore, the effect can not be discounted.

Critical Habitat

The determination for critical habitat modification is based on results for aquatic and terrestrial plants as well as the alteration in habitat due to the effect of reduction in prey.
There were no LOCs exceeded for aquatic vascular or non-vascular plants. There was no effect to any terrestrial plants at the highest concentration tested.

However, due to the effects of Ziram on prey items resulting in alteration of habitat there is a 'habitat modification' determination. Based on probit results for acute effects for the potential for significant chance of individual effects for fish and aquatic invertebrates there is a "habitat modification" determination for aquatic-phase PCEs.

Based on probit results for acute effects for the potential for significant chance of individual effects for frogs, terrestrial invertebrates, and small mammals there is a "habitat modification" determination for terrestrial-phase PCEs.

Determinations for direct and indirect effects of Ziram to the CRLF are presented in **Error! Reference source not found.**. Determinations for habitat modification are presented in **Error! Reference source not found.**. Given the LAA determination for the CRLF and potential modification of designated critical habitat, a description of the baseline status and cumulative effects for the CRLF is provided in Attachment 2.

Table 34 Effects Determination Summary for Ziram Use and the CRLF**.**

Assessment Endpoint	Effects Determination [1]	Basis for Determination
Survival, growth,		**Potential for Direct Effects**

Table 34 Effects Determination Summary for Ziram Use and the CRLF**.**

Assessment Endpoint	Effects Determination [1]	Basis for Determination
and/or reproduction of CRLF individuals	LAA[1]	***Aquatic-phase (Eggs, Larvae, and Adults)***: For acute aquatic-phase direct effects, the May affect is based on all modeled uses RQs (almonds, apples/pears, apricots, cherries, grapes, nectarines, peaches, pecans, wine grapes) exceeding the LOC using the fish as a surrogate for the CRLF. There was an LAA determination based on the lines of evidence including the probit analysis to calculate the probability of individual effects.
		Terrestrial-phase (Juveniles and Adults): For acute and chronic terrestrial-phase direct effects, the "may affect" is based on all acute and chronic modeled uses T-REX RQs exceeding the LOC. T-HERPs acute dose-based RQs exceeded the LOC for all modeled uses (almonds, apples/pears, apricots, cherries, grapes, peaches and pecans). The LAA determination for acute direct effects using the bird as a surrogate resulted from lines of evidence for T-HERPS RQs exceeding the LOC for all use scenarios and the probability analysis resulting estimating the chance of individual effects.
		A refinement following the "May Affect" determination for chronic effects using T-HERPs dose-based RQs also exceeded the LOC for all modeled uses (almonds, apples/pears, apricots, cherries, grapes, peaches and pecans) for chronic effects.
		Potential for Indirect Effects
		Aquatic prey items, aquatic habitat, cover and/or primary productivity *Aquatic Prey:* There was a "may affect" determination for aquatic invertebrates based on LOC exceedences for all use scenarios for acute effects to the CRLF. The "LAA" determination was based on the lines of evidence including probit analysis indicating significant effects for individuals for almonds, apples, apricots, grapes, nectarines, pears, peaches, pecans and wine grapes for both Ziram and the degradate Thiram.
		Aquatic habitat: The indirect habitat effect of Ziram on the CRLF is based on an analysis of the acute effects of aquatic invertebrates, there was a "may affect" based on the LOC exceedence for all use scenarios. There was a significant effect indicated by the probit analysis for all use scenarios for uses except cherries.
		Terrestrial prey items, riparian habitat The RQs for all use scenarios exceeded the LOC threshold (0.05) for terrestrial invertebrates for Ziram. The LAA determination is based on lines-of-evidence including a significant individual effect for all use scenarios for Thiram.
		Acute RQs for fish as prey items exceed LOCS for all uses. There was an "LAA" determination using the Probit analysis as a refinement based on significant chance of individual effects for all uses.

Table 34 Effects Determination Summary for Ziram Use and the CRLF**.**

Assessment Endpoint	Effects Determination [1]	Basis for Determination
		The T-REX acute and chronic analysis for mammals as prey resulted in a "may affect" Lines of evidence for acute effects on mammals as prey items include RQs exceeding the acute risk LOC=0.5 and the probit analysis resulting in significant chance of individual effects resulting in an LAA determination.
		The T-REX analysis for chronic effects of mammals as prey items resulted in RQs exceeding the LOC for all uses. There is an overlap between areas of the expected adverse affect and where the species is located; therefore, the effect can not be discounted.
		Riparian Habitat Effects to riparian habitat were based on alteration in habitat due to reduction in prey. There is a significant reduction in prey for frog using the fish as a surrogate due to RQs exceeding the acute risk LOC = 0.5 for all uses.
		For acute effects, there is a significant reduction in prey for mammals due to RQs exceeding the acute risk LOC = 0.5 for all uses for both Ziram and Thiram..

[1] May affect, likely to adversely affect (LAA)

Table 37. Effects Determination Summary for Ziram Use and CRLF Critical Habitat Impact Analysis.

Assessment Endpoint	Effects Determination [1]	Basis for Determination
Modification of aquatic-phase PCE	Habitat Modification[1]	Aquatic PCEs are based on the effect of Ziram on aquatic and terrestrial plants, as well as the effects on fish and aquatic invertebrates consumed by the CRLF. Although there were no effects on aquatic or terrestrial plants, there were effects on fish and aquatic invertebrates consumed by the CRLF. Based on potential effects for both fish and aquatic invertebrates based on LOC exceedence and the chance of individual effects for all uses there is a habitat modification determination for Ziram.
Modification of terrestrial-phase PCE		Terrestrial PCEs are based on the effect of Ziram on terrestrial plants, as well as the effects on prey consumed by the CRL.F. Although there were no effects on terrestrial plants, there were effects on terrestrial invertebrates, mammals and frogs consumed by the CRLF. Ziram RQs for the bee exceeded the LOC (0.05) for all use scenarios. The more sensitive Thiram RQs for the honey bee exceeded the LOC (0.05) for all use scenarios. The probit analysis indicated significant chance of individual effects for all uses.
		T-REX RQs exceeded the LOC for all use scenarios for acute and chronic dietary effects for small mammals. In addition the probit analysis indicated significant effects for all uses for both Ziram and Thiram.

		Using the bird as a surrogate for frogs, all modeled uses resulted in T-REX RQs exceeding the LOC=0.1. Lines of evidence, including - probit analysis indicated an "LAA: determination based on significant chance of individual effect for all uses.

[1] Habitat Modification

Based on the conclusions of this assessment, a formal consultation with the U. S. Fish and Wildlife Service under Section 7 of the Endangered Species Act should be initiated to seek concurrence with the LAA determinations and to determine whether there are reasonable and prudent alternatives and/or measures to reduce and/or eliminate potential incidental take.

When evaluating the significance of this risk assessment's direct/indirect and adverse habitat modification effects determinations, it is important to note that pesticide exposures and predicted risks to the species and its resources (i.e., food and habitat) are not expected to be uniform across the action area. In fact, given the assumptions of drift and downstream transport (i.e., attenuation with distance), pesticide exposure and associated risks to the species and its resources are expected to decrease with increasing distance away from the treated field or site of application. Evaluation of the implication of this non-uniform distribution of risk to the species would require information and assessment techniques that are not currently available. Examples of such information and methodology required for this type of analysis would include the following:

- Enhanced information on the density and distribution of CRLF life stages within specific recovery units and/or designated critical habitat within the action area. This information would allow for quantitative extrapolation of the present risk assessment's predictions of individual effects to the proportion of the population extant within geographical areas where those effects are predicted. Furthermore, such population information would allow for a more comprehensive evaluation of the significance of potential resource impairment to individuals of the species.
- Quantitative information on prey base requirements for individual aquatic- and terrestrial-phase frogs. While existing information provides a preliminary picture of the types of food sources utilized by the frog, it does not establish minimal requirements to sustain healthy individuals at varying life stages. Such information could be used to establish biologically relevant thresholds of effects on the prey base, and ultimately establish geographical limits to those effects. This information could be used together with the density data discussed above to characterize the likelihood of adverse effects to individuals.
- Information on population responses of prey base organisms to the pesticide. Currently, methodologies are limited to predicting exposures and likely levels of direct mortality, growth or reproductive impairment immediately following exposure to the pesticide. The degree to which repeated exposure events and the inherent demographic characteristics of the prey population play into the extent to which prey resources may recover is not predictable. An enhanced understanding of long-term prey responses to pesticide exposure would allow for a more refined determination of the magnitude and duration of resource impairment, and

together with the information described above, a more complete prediction of effects to individual frogs and potential modification to critical habitat.

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